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Original Articles

THE STABILITY OF THE REAGIN-NEUTRALIZING SUBSTANCE IN RAGWEED POLLEN AT VARIOUS HYDROGEN-ION CONCENTRATIONS

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THE conditions influencing the stability of the biologically active material in extracts of ragweed pollen are of considerable importance both in their bearing on the choice of the most suitable conditions for extraction of the pollen and storage of the extracts and in relation to the chemical manipulations that have been employed in numerous attempts to isolate and to characterize chemically the ragweed atopen. In an investigation of the effect of temperature on the ragweed atopen, Arbesman and Eagle[1] have demonstrated that the reagin-neutralizing activity of ragweed pollen extracts is destroyed by relatively short exposure to a temperature of 56° C. Since many thermolabile biologic materials are known to be inactivated or altered also by slight excesses of acid or alkali, it seemed advisable to investigate the effect of the pH of the extract upon the stability of the ragweed atopen. The present investigation was therefore undertaken to determine the stability of the active material in extracts of ragweed pollen at various hydrogen-ion concentrations.

The reported data on this question are fragmentary and inconclusive. The results reported by Gay[2] indicate that the extracts are equally stable at the pH values of 4.0, 7.4, and 8.4, and the work of Unger and Moore[3] suggests that ragweed pollen extracts are equally stable between pH 6.4 and 7.5. Stull, Chobot, and Cooke,[4] and Coca and Milford,[5] without giving experimental data, state that slightly acid extracts are less stable than slightly alkaline ones, while Grove and Coca[6] have observed a slow deterioration at pH 7.0. In a preliminary report, Cooke and his co-workers[7] obtained satisfactory therapeutic results with heated alkaline saline extracts, although they mentioned that the reagin-

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neutralizing power was less than that of an untreated saline extract. In a more recent communication,[8] they were unable to verify their earlier favorable therapeutic results. Brown and co-workers[9] stated that the ragweed atopen is completely inactivated by the presence of 0.1 N NaOH.

As will be shown in the present paper, ragweed pollen extracts are stable at 37° C. only within a well-defined and relatively narrow pH zone (5.0 to 7.0). The reagin-neutralizing activity of such extracts is almost completely destroyed within a relatively short time on either side of the zone of optimum stability.

EXPERIMENTAL DATA

The extracts were all of whole, short ragweed pollen. Previous treatment with organic solvents to remove lipoidal material was avoided because of the possibility that such treatment might alter the properties of the proteins present in the pollen. For Experiment 1, 8.0 Gm. of pollen were extracted overnight with 200 c.c. of unbuffered 0.85 per cent NaCl solution. The pollen residue was then filtered off and washed with 80 c.c. of saline solution. The combined washings and opalescent extract were employed without further treatment. The extract for Experiment 2 was prepared by overnight extraction of 4.0 Gm. of pollen with 100 c.c. of saline solution and removal of the residue, without washing, by centrifugation. The extracts used in Experiments 3, 4, and 5 were similiar to that employed for Experiment 2 with the exception that they were passed through a Berkefeld filter to remove the finely particulate matter which was not thrown down by an ordinary laboratory centrifuge. The pH of the extracts was approximately 5.5 to 6.0.

Ten cubic centimeter aliquots of the extracts were adjusted to the desired pH values by the addition of 0.1 N HCl or NaOH, the pH being determined on a Beckman pH meter. In order to increase the buffering capacity of some of the aliquots, a 0.2 M solution of KH_2PO_4 was added, prior to the pH determination, in amounts sufficient to give a phosphate concentration of approximately 0.03 M.

The aliquots were then placed in a 37° C. air incubator for four hours, after which the aliquots were adjusted to a pH of 5.5 to 6.0, approximately that of the original extracts. The solutions were then sterilized by Berkefeld filtration and stored at –10° C. until their reagin-neutralizing power was determined.

The reagin-neutralizing activity of the solutions was determined by the method recently described by Arbesman and Eagle.[10] Briefly, this method consists of the determination of the minimal amount of the extract that suffices to neutralize the skin-sensitizing antibody present in the serum of ragweed-sensitive persons. The presence of free, unneutralized reagin was demonstrated by injecting the various extract-serum mixtures into the skin of patients who were nonsensitive to ragweed pollen. Each of the sites was retested twenty-four hours later by the injection of approximately 0.05 c.c. of a 1:200 dilution of a standard

TABLE I

ILLUSTRATIVE PROTOCOL OF THE BIOLOGIC ASSAY (EXPERIMENT 3)

pH	3.74	4.09	4.49	7.16	7.43	7.70	CONTROL	3.74	4.09	4.49	7.16	7.43	7.70	CONTROL
DILUTION OF EXTRACT ADDED TO EQUAL VOLUME OF 1:5 DILUTION OF SERUM G.T.	SKIN REACTION OBSERVED UPON RETESTING SENSITIZED SITE WITH 1:200 DILUTION OF UNTREATED EXTRACT IN													
	RECIPIENT NO. 22							RECIPIENT NO. 23						
whole	2-10	0-0	0-0		0-0	0-0		2-10	0-0	0-0		0-0	0-0	
1:5	2-10	0-0	0-0		0-0	0-0		2-12	0-0	0-0		0-0	0-0	
1:10	3-20	2-12	0-0	0-0	0-0	2-7		2-10	2-10	0-0	0-0	0-0	2-7	
1:15	5-30	3-10	0-0	0-0	0-0	2-10		2-15	3-15	0-0	0-0	0-0	2-10	
1:20	5-30	3-20	2-11	0-0	0-0	2-10	0-0	3-20	3-15	2-6	0-0	0-0	2-20	0-0
1:30		3-20	2-15	0-0	2-10		0-0		3-15	2-6	0-0	2-10		0-0
1:40			3-20	2-10	2-15		0-0			2-18	2-10	2-15		0-0
1:60				2-12			0-0				2-12			0-0
1:80							0-0							0-0
1:120							2-8							2-10
Minimal neutralizing dilution	<1:1	1:5	1:15	1:30	1:20	1:5	1:80	<1:1	1:5	1:15	1:30	1:20	1:5	1:80
Dilution by reagents	1:1.57	1:1.54	1:1.49	1:1.6	1:1.6	1:1.62	none	1:1.57	1:1.54	1:1.49	1:1.6	1:1.6	1:1.62	None
Net neutralizing dilution	<1:1.57	1:7.7	1:22	1:48	1:32	1:8.1	1:80	<1:1.57	1:7.7	1:22	1:48	1:32	1:8.1	1:80
Per cent activity	<2	10	28	60	40	10	100	<2	10	28	60	40	10	100

Equal volumes of sensitive serum G. T., diluted 1:5, and of serial dilutions of the extract to be assayed were mixed. After one hour at 37° C., 0.1 c.c. portions of these mixtures were injected into sites on the volar aspect of the arms of nonsensitive patients. Sites were retested twenty-four hours later for presence or absence of free reagin by injection of 0.05 c.c. of 1:200 untreated pollen extract. In the table, the skin reactions that were observed are expressed as size in millimeters of the wheal and erythema, respectively.

These particular results are indicated in Fig. 1 by open squares.

(untreated) extract. At least two normal persons were used for each assay. In general, as is illustrated in Table I, the relative activities of a series of extracts as determined in two recipients gave satisfactorily concordant results. As pointed out by Arbesman and Eagle, this method gives the reagin-neutralizing power of an extract *relative* to that of a second extract and does not give the *absolute* neutralizing activity. Accordingly, the neutralizing power of the solutions incubated at various pH values was compared in all cases to that of an untreated aliquot of the same extract that had been kept at –10° C. throughout the course of the experiment and that had, therefore, probably suffered no significant loss in activity. The activity of each of the experimental solutions is expressed in Table I and Fig. 1 as per cent of the activity of the corresponding untreated control.

RESULTS

For the sake of brevity and to avoid unnecessary repetition of experimental results, the complete protocol of only one experiment (Experiment 3) is given in Table I. The results of this particular experiment are indicated in Fig. 1 by open squares. The pH values at the head of the table are the means of the values that were obtained before and after incubation. These two values did not differ significantly. At the left of the table are given the dilutions of the incubated and neutralized extract that were used in the biologic assay. Since these dilutions do not take into account the reagents added to the original aliquots, they are corrected accordingly to give the "net neutralizing dilution" as indicated at the bottom of the table. The size of the skin reactions given in the body of the table is expressed as the diameter in millimeters of the wheal and of the erythema, the smaller number of the two referring in each case to the wheal.

A graphical summary of all of the experimental work is presented in Fig. 1. In order to make the distribution of the points on the graph conform more closely to the experimental errors in assaying the activity of the solutions, it has been found convenient to plot the square root of the per cent activity vs. the pH. The lowest value on the ordinate has been termed 0 to 1 per cent, traces of activity of this magnitude not being detected by the method of assay that was employed.

From the graph it is apparent that at 37° C. 50 per cent or more of the activity is destroyed in four hours in solutions more acid than pH 5.0 or more alkaline than pH 7.0 and that only 25 per cent or less of the activity remains in solutions outside the pH limits of approximately 4.5 and 7.5. The pH region of maximum stability lies between the approximate values of 5.5 and 6.5.

In all of the solutions a precipitate, the chemical nature of which has not as yet been investigated, developed during the period of incubation or shortly after neutralization of the aliquot. It was quite voluminous

in the solutions for Experiments 1 and 2 and in these cases appeared to be due, at least in part, to an aggregation of the microscopic particles not removed by centrifugation. However, precipitates, less in amount, also developed in the solutions of Experiment 3, when extracts were used which had been passed through a Berkefeld filter and which contained no microscopically visible particles. The amount of precipitate was greatest in those aliquots with pH farthest removed from the region of maximum stability. In order to control the possibility that the active principle of the extracts was carried down in the precipitate in an

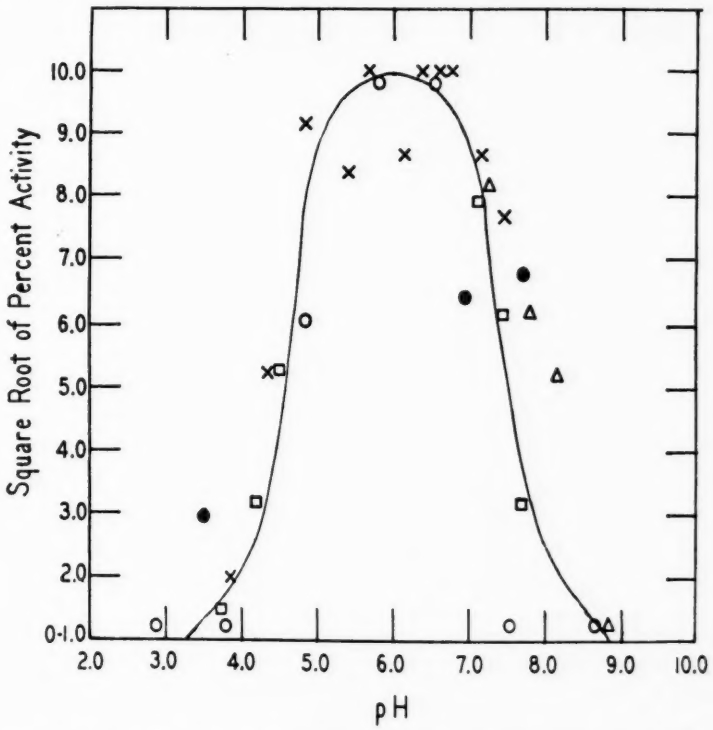

Fig. 1.—The effect of pH on the activity of short ragweed pollen extracts incubated for four hours at 37° C. The activity is expressed as per cent of the activity of the corresponding untreated controls. The several experiments are denoted as follows: 1, ○; 2, ×; 3, □; 4, ●; 5, △.

active form, and removed when the solution was sterilized by Berkefeld filtration, in Experiments 4 and 5 the Berkefeld filtration after the incubation was omitted. The original, centrifuged extracts were filtered through sterile Berkefeld filters, and the experiments were conducted with sterile reagents and containers throughout. After neutralization, the entire experimental mixtures, including the precipitates, were used in the assays of biologic activity. As is shown in Fig. 1, the results of these two experiments agree very well with those of the previous experiments, indicating that the observed loss of activity was not due to the formation of a biologically active precipitate.

DISCUSSION

There is an interesting similarity between the general shape of the present curve and that of the curve obtained by Bull and Neurath[11] for the effects of pH on the denaturation of egg albumin by mechanical shaking. That the hydrogen-ion concentration also is a determining factor in the rate of thermal denaturation of proteins has been shown by the experiments of Lewis[12] with oxyhemoglobin and egg albumin. Lewis also showed that the pH of slowest thermal denaturation does not necessarily coincide with the isoelectric point of the protein, but lay, in the cases he studied, slightly on the acid side of neutrality. The present results are therefore consistent with those that might be expected if the inactivation of the pollen atopen were fundamentally a process of protein denaturation. However, a definite conclusion as to whether or not this is the case must await conclusive evidence of the protein nature of the biologically active material in ragweed pollen.

SUMMARY

The effect of pH on the inactivation of ragweed pollen extracts at 37° C. has been determined.

After four hours, more than 75 per cent of the activity is destroyed in solutions the pH of which lies outside the approximate limits of 4.5 and 7.5. The pH region of maximum stability has been found to be approximately 5.5 to 6.5.

It is tentatively suggested that the inactivation may be due to the denaturation of an active protein component of ragweed pollen.

REFERENCES

1. Arbesman, C. E., and Eagle, H.: The Thermolability of Ragweed Pollen Extract and Its Corresponding Reagin, J. ALLERGY **11:** 18, 1939.
2. Gay, L. N.: The Effect of Heat and Hydrogen-Ion Concentrations on the Extract of Short Ragweed, J. Immunol. **11:** 371, 1926.
3. Unger, L., and Moore, M. B.: The Effects of Varying Hydrogen-Ion Concentration Upon the Stability of Pollen Extracts, J. ALLERGY **8:** 22, 1936.
4. Stull, A., Chobot, R., and Cooke, R. A.: Chemical and Clinical Studies on Pollen (Preliminary Report), J. ALLERGY **1:** 470, 1930.
5. Coca, A. F., and Milford, E. L.: Studies on Specific Hypersensitiveness. The Preparation of Fluid Extracts and Solutions for Use in the Diagnosis and Treatment of Atopic Conditions, J. Immunol. **10:** 555, 1925.
6. Grove, E. F., and Coca, A. F.: Studies in Specific Hypersensitiveness. On the Nature of the Atopens of Pollens, House Dust, Horse Dander, and Green Pea, J. Immunol. **10:** 471, 1925.
7. Cooke, R. A., Stull, A., Hebald, S., Loveless, M., and Sherman, W. B.: A Preliminary Report on the Preparation and Use of Modified (Denatured) Extracts in the Treatment of Allergic Conditions, J. ALLERGY **8:** 278, 1937.
8. Stull, A., Cooke, R. A., Sherman, W. B., Hebald, S., and Hampton, S. F.: Experimental and Clinical Study of Fresh and Modified Pollen Extracts, J. ALLERGY **11:** 439, 1940.
9. Brown, A., Milford, E. L., and Coca, A. F.: Studies on Contact Dermatitis. The Nature and Etiology of Pollen Dermatitis, J. ALLERGY **2:** 301, 1931.
10. Arbesman, C. E., and Eagle, H.: The Assay of Ragweed Pollen Extracts, J. ALLERGY **10:** 521, 1939.
11. Bull, H. B., and Neurath, H.: The Denaturation and Hydration of Proteins. Surface Denaturation of Egg Albumin, J. Biol. Chem. **118:** 163, 1937.
12. Lewis, P. S.: The Kinetics of Protein Denaturation, Biochem. J. **20:** 965, 978, 1926.

SPECIES SPECIFICITY OF ALTERNARIA IN ASTHMA AND HAY FEVER

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THERE is a paucity of literature on the specificity of mold spores acting as atopic excitants. Sulzberger[1] has reported an instance of *Trichophyton* hypersensitivity in which reagins were present for two species of *Trichophyton* but none for *Oidiomycin*. This suggests genus specificity but lack of species specificity. Persons and Martin[2] studied a patient with a dermatosis whose serum showed reagins for six saprophytic fungi cultured from the lesion. In vivo and in vitro cross neutralizations were complete with three of these: *Cladosporium herbarum, Nigrospora* species, and *Aspergillus fumigatus*. In this instance even genus specificity was lacking.

Rackemann and his co-workers[3-5] reported the cases of four greenhouse workers who had asthma as a result of contact with *Cladosporium fulvum,* Cooke, which grows on tomato vines during certain seasons. All of these patients showed a highly specific skin reaction to this mold and responded weakly or not at all to other species of *Cladosporium*. This specificity is probably the result of a tremendous exposure to a single species of mold. Lamson and Rogers[6] also failed to discover a common antigenic factor in four species of *Aspergillus* during routine skin testing of patients in southern California.

Almon and Stovall[7] have reviewed the literature on the experimental production of antisera to fungi. In most of this work, pathogens of the *Monilia* and related yeast groups have been investigated. It is clear that crossed reactions between related species and even genera as demonstrated by agglutination, precipitation, and complement fixation are common. When absorption of precipitin on the intact mycotic bodies was undertaken by Kesten and his co-workers,[8] a greater degree of specificity was evident by precipitation with polysaccharides of several strains of *Monilia*. It must be emphasized that these experiments were conducted under the artificial circumstances of producing rabbit antisera by injection of antigen, whereas the problem under consideration deals with reaginic sera of atopic humans. In all the experimental work with animals, one fact stands out clearly: fungi do not produce the highly specific immunologic responses characteristic of pathogenic bacteria.

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In view of the increasing importance attached to mold spore hypersensitivity in human respiratory allergy, the present study was undertaken to determine whether species specificity commonly exists in a single genus of mold. If such is the case, it would be necessary to test each patient with a vast number of species of each common genus. If, however, the various genera are relatively homogeneous in so far as the human respiratory mucous membrane is concerned, the problem of skin testing is greatly simplified. *Alternaria* was chosen for investigation because it is widely accepted as a common cause of atopic reactions and because an ample supply of reaginic serum was available. The method consisted of direct testing of patients with different species of Alternaria and in vivo and in vitro neutralization experiments with the sera of these patients.

DIRECT TESTING

Method.—At the Children's Hospital, twenty-eight patients, all thought to be clinically sensitive to *Alternaria,* were tested by the intracutaneous method with four species of *Alternaria,* as follows: (1) *Alternaria* sp. from carnation blight, (2) *Alternaria* sp. from dying geranium, (3) *Alternaria* sp. from decaying squash, and (4) *Alternaria* sp. commonly occurring in the air in Boston. The first three species were supplied by the Massachusetts State Agricultural College Experiment Station through the courtesy of Dr. E. F. Guba.

The extracts were made from the ground mats grown on potato-dextrose medium and extracted with alkaline extracting fluid. After passage through a Seitz filter, they were standardized to 0.1 mg. total nitrogen per cubic centimeter.

Through the courtesy of Dr. F. M. Rackemann and with the cooperation of Dr. T. G. Randolph, thirteen adult patients at the Massachusetts General Hospital were tested intracutaneously to six species as follows: *Alternaria circinans, Alternaria solani, Alternaria geophila, Alternaria radicina, Alternaria mali,* and *Alternaria iridis.* Although their skins reacted to *Alternaria,* the patients were not necessarily clinically sensitive. These species were chosen from sixteen supplied by the American Type Culture Collection at Washington because they sporulated readily on potato-dextrose agar. They were extracted with alkaline extracting fluid, but they were not standardized by nitrogen determinations.

Results.—Twenty-seven of the twenty-eight children tested with four species of *Alternaria* reacted to all species but usually failed to react to other genera, e.g., *Hormodendron, Aspergillus,* and *Penicillium.* In one instance the patient reacted to three species but failed repeatedly to respond to the fourth. The tests were moderately uniform in size, usually producing a pseudopodic wheal 10 to 15 mm. in diameter. There

was a tendency for the *Alternaria* sp. obtained from decaying squash to produce slightly larger reactions. This was thought to be due to the more abundant spores in the mats used for making the extract. From these tests it would appear that the patients were sensitive to a common factor in these four species of *Alternaria*.

The results in the thirteen adult patients tested at the Massachusetts General Hospital with six different species are not so definite. Seven subjects reacted strongly to all six species; three reacted to five species; one, to three species; and two, to only two species. However, those responding to all six species gave large reactions, while those reacting to five or fewer species responded with smaller reactions. Furthermore, clinical sensitivity was doubtful. These facts tend to corroborate the homogeneity of the *Alternaria* genus but do not entirely confirm it.

DESENSITIZATION OF PASSIVELY SENSITIZED SITES

Method.—Sera from six untreated patients suffering from asthma or hay fever and clinically sensitive to *Alternaria* were used to prepare duplicate passively sensitized sites by injecting 0.1 c.c. of each into the skin of nonatopic recipients. On the next day, when the reagins were fixed in the tissues, the first site was tested with 0.01 c.c. of *Alternaria* sp. A (see below) and then retested daily until no reaction occurred. The following day the site was tested to *Alternaria* sp. B. If no reaction occurred, the site was tested with an unrelated antigen to which the donor of the serum was known to be sensitive to prove that the area was still capable of reacting. Conversely, the second site was desensitized to *Alternaria* sp. B and then tested with *Alternaria* sp. A and later with the unrelated antigen.

The two species, A and B, used in this experiment were commonly found in the air in Boston. They differed culturally in that the growth of *Alternaria* sp. A produced a dark brown, powdery surface of spores on solid Sabouraud's medium, whereas the surface of sp. B consisted of a loose, white mesh of mycelium in which spores were found to be less abundant. The extracts were made from the entire mat grown on the liquid Sabouraud's medium. No effort was made to standardize the extracts since it is not known whether the active nitrogen is in the spore or mycelium. On direct testing, the extract of sp. A usually gave slightly larger reactions than that of sp. B, perhaps because it was richer in spore nitrogen.

Results.—With two of the six sera cross neutralization was complete between *Alternaria* sp. A and sp. B. An example of one such experiment is shown in Fig. 1. In all six instances, sp. A completely neutralized the reagins to sp. B, whereas sp. B failed once to neutralize sp. A and three times only partly neutralized sp. A. Fig. 2 illustrates the latter situation. It would appear that sp. A either contained an

antigen not present in sp. B or that its extract was richer in atopic excitant. The latter seems to be the more reasonable explanation because direct reactions with sp. A were, in general, somewhat greater and because the mats from which the extracts were made were much richer in spores. Furthermore, fewer treatments of sensitized sites with sp. A were necessary to desensitize the site than was true with the less powerful sp. B extract.

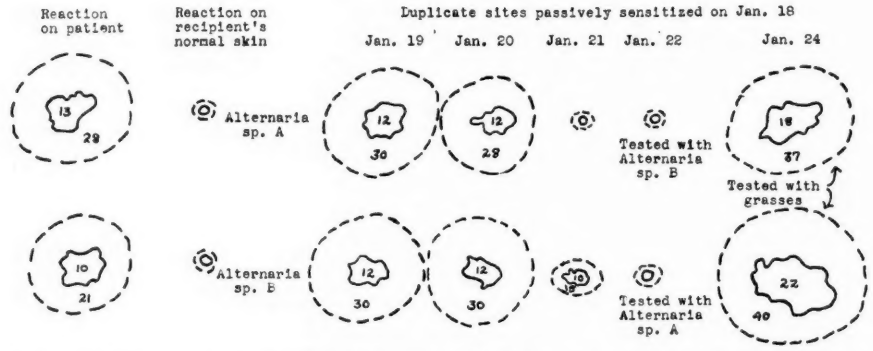

Fig. 1.—Desensitization of passively sensitized sites. Complete cross neutralization between *Alternaria* sp. A and sp. B.

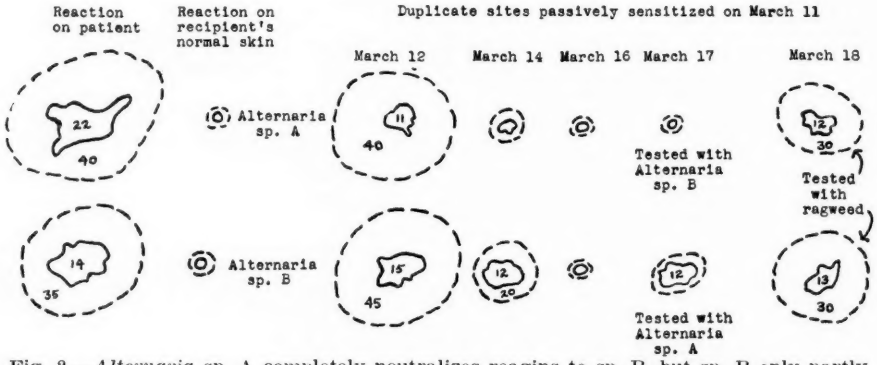

Fig. 2.—*Alternaria* sp. A completely neutralizes reagins to sp. B, but sp. B only partly neutralizes sp. A.

IN VITRO NEUTRALIZATION

Method.—The extracts were the same as those used for direct testing in the Children's Hospital, namely, *Alternaria* species from carnation blight, dying geranium, and decaying squash. Since these arise from different biologic sources and are morphologically different, they are probably different species. Sera were taken from fourteen untreated children clinically sensitive to *Alternaria.* Mixtures of two parts extract to one part serum were made for each species under sterile precautions and were allowed to stand for twenty-four hours in the icebox. Each mixture was then injected in 0.1 c.c. amounts in triplicate sites on non-

atopic recipients. Triplicate control sites of two parts physiologic solution of sodium chloride and one part serum were also prepared. After twenty-four hours, each series of sites was tested with one of the three species of *Alternaria*. Fig. 3 is an example of one experiment.

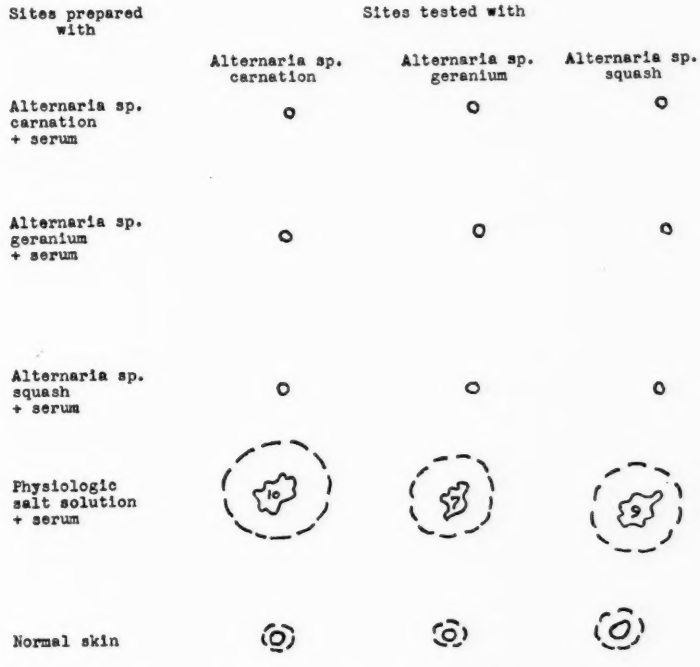

Fig. 3.—Complete cross neutralization in vitro between three species of *Alternaria* from different biologic sources.

TABLE I

SUMMARY OF FOURTEEN NEUTRALIZATION EXPERIMENTS

SERUM NEUTRALIZED WITH *Alternaria* SP. FROM	SITES TESTED WITH *Alternaria* SP. FROM	DEGREE OF NEUTRALIZATION		
		COMPLETE	PARTIAL	NONE
Carnation	Geranium	9	5	0
Carnation	Squash	1	5	7
Geranium	Carnation	9	5	0
Geranium	Squash	1	4	5
Squash	Carnation	14	0	0
Squash	Geranium	14	0	0

Results.—It is a striking fact that all fourteen sera contained strong reagins to all three species of *Alternaria*. In only one instance, however, was complete cross neutralization between the three species demonstrated (Fig. 3). The extract of *Alternaria* obtained from decaying squash completely neutralized the reagins to the other two species without exception. On the other hand, the *Alternaria* obtained from carna-

tion blight and from dying geranium tended to neutralize each other but failed in seven and five instances respectively even partly to neutralize the reagins to *Alternaria* sp. squash. Fig. 4 illustrates the most frequent result, and Table I summarizes the fourteen experiments.

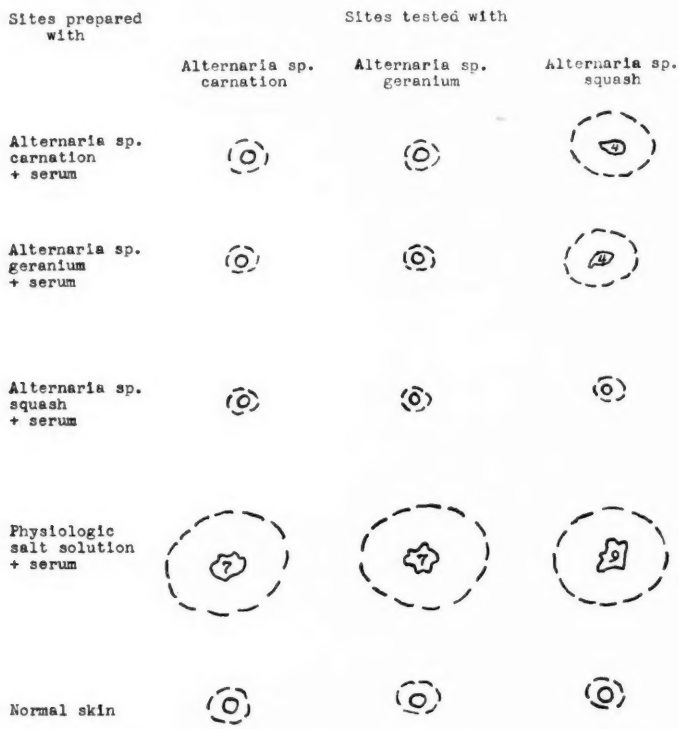

Fig. 4.—Complete neutralization of all reagins by *Alternaria* sp. squash. Complete cross neutralization between species carnation and species geranium. Partial neutralization of species squash by species carnation and species geranium.

DISCUSSION

The direct testing work strongly points to a common atopen in all species of *Alternaria* studied. The in vivo and in vitro neutralization experiments are not conclusive, although they indicate a frequent but not uniform tendency to cross neutralization. One fact stands out clearly: in both sets of experiments, one species (*Alternaria* sp. A and *Alternaria* sp. squash) invariably neutralized the reagins to all others tested. The extracts of both these species, although similar in nitrogen content, usually gave larger reactions on direct testing than the other species (*Alternaria* sp. B, sp. carnation, and sp. geranium). These facts would suggest either that these species contained an atopen not present in the others or that they were richer in the common atopic excitant. The latter explanation gains credence from the fact that these two stronger extracts were derived from species which produce more abundant spores.

It seems likely, but has not yet been proved, that the spores are the important part of the mold in producing respiratory allergy. If this is so, an extract made primarily of spores would be much more powerful than one made from a mat poor in spores even if the nitrogen contents were the same. Investigations which are being carried out at the present time would indicate that this is true. Therefore, one must question the significance of an experiment in which an extract made from a mold poor in spores fails to neutralize reagins. On the positive side of the argument is the fact that the extracts made from cultures rich in spores invariably neutralized all reagins to all species. Therefore, the neutralization experiments tend to confirm the impression gained from direct testing, namely, that the genus *Alternaria* is homogeneous in specificity in relation to human atopy. However, the final answer cannot be given until methods are available for standardizing the atopic content of *Alternaria* extracts.

SUMMARY

1. Direct testing of children and adults with several species of *Alternaria* varying in cultural characteristics and biologic sources indicates a common atopic excitant in the genus *Alternaria*.

2. This result is only partly confirmed by in vivo and in vitro neutralization experiments.

3. Reciprocal neutralization was common but not invariable.

4. This contradiction may be explained partly by the lack of a method for accurate standardization of the atopic content of mold extracts.

5. These experiments suggest, but do not prove, that the genus *Alternaria* is relatively homogeneous in relation to human atopic immunology.

REFERENCES

1. Sulzberger, M. B.: Experiments in Passive Transference of Urticarial Hypersensitiveness to Fungous Extracts, J. Immunol. **23**: 73, 1932.
2. Persons, E. L., and Martin, D. S.: Passive Transfer Antibodies for Six Saprophytic Fungi in a Patient With a Superficial Scaling Dermatosis, J. Clin. Investigation **15**: 429, 1936.
3. Rackemann, F. M., Randolph, T. G., and Guba, E. F.: The Specificity of Fungous Allergy, J. ALLERGY **9**: 447, 1938.
4. Guba, E. F., and Rackemann, F. M.: Species of Cladosporium on Tomato and the Allergic Response in Man as an Aid to Their Identification, Mycologia **30**: 625, 1938.
5. Rackemann, F. M.: Molds as a Cause of Hay Fever and Asthma, Tr. Am. Clin. & Climatol. A. **53**: 194, 1939.
6. Lamson, R. W., and Rogers, E. L.: Skin Hypersensitivity to Molds, J. ALLERGY **7**: 582, 1936.
7. Almon, L., and Stovall, W. D.: Serologic Reactions of Cultures of Monilia and of Some Other Yeast-Like Fungi, J. Infect. Dis. **55**: 12, 1934.
8. Kesten, H. D., Cook, D. H., Mott, E., and Jobling, J. W.: Specific Polysaccharides From Fungi, J. Exper. Med. **52**: 813, 1930.

STUDIES IN ABSORPTION OF UNDIGESTED PROTEIN

X. Absorption From the Gall Bladder

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IN PREVIOUS investigations in the human and in the *Rhesus* monkey it has been shown that the absorption of unaltered protein occurs from such mucous membrane surfaces as esophagus (monkey),[1] stomach (monkey),[1] small intestine (human),[2] colon (human),[3] and rectum (human).[4] These studies have demonstrated that the absorption phenomenon in the *Rhesus* monkey is analogous to that occurring in the human. Obviously, experimental studies on the absorption of unaltered protein from the gall bladder are not feasible in humans. The present experiments were, therefore, designed to study this question in the *Rhesus* monkey.

TECHNIQUE

The technique described by Straus and Walzer[5] in a previous communication was employed in these experiments. It involved the sensitization of a cutaneous site on a *Rhesus* monkey by the intracutaneous injection of a human serum containing skin-sensitizing antibodies (atopic reagins) for the antigen to be studied. After an interval of forty-eight hours to permit fixation of the antibodies at the cutaneous site, the specifically related antigen was introduced directly into the lumen of the gall bladder by means of needle and syringe. An urticarial reaction, occurring at the sensitized cutaneous site, marked the entrance of the unaltered antigen into the circulation.

The *sensitizing serum* was obtained from an asthmatic patient, "P," who evidenced marked positive reactions to intracutaneous tests with dilute cottonseed extract. This serum showed an exceptionally high titer in atopic reagin content for cottonseed protein (1 to 1,024 by the dilution method of Coca and Grove[6]).

Food was withheld from the animal for at least twenty-four hours previous to the experiment, but small amounts of water were given. The *protein test meal* consisted of a cottonseed "milk," which was prepared by mixing 10 Gm. of raw ground cottonseed in 30 ml. of tap water. The *absorption time* of each animal was represented by the

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interval which elapsed between the administration of the test meal and the onset of the reaction at the sensitized cutaneous site.

EXPERIMENTS

Experiment 1.—The animal employed in this study was a male monkey, weighing 2.5 kg. The details of the experiment, in chronological order, follow:

Jan. 28, 1938. 9:00 A.M.: A cutaneous site on each thigh was passively sensitized to cottenseed antigen with an intracutaneous injection of 0.05 ml. of "P" serum.

Jan. 30, 1938. 9:35 A.M.: The intravenous injection of 1.0 grain of veterinary nembutal resulted in immediate narcosis.

9:45 A.M.: The abdomen was opened. The biliary tract was exposed. The common duct was clamped just above its entrance into the duodenum.

10:13 A.M.: One milliliter of bile was withdrawn from the gall bladder by syringe, and the needle was left in situ.

10:14 A.M.: Two milliliters of cottonseed "milk" were injected into the lumen of the gall bladder by syringe through the needle left in situ. There was no evidence of bleeding at the site of puncture after the needle was withdrawn.

10:20 A.M.: Wheal formation began to develop at the sensitized cutaneous sites. This indicated the absorption of the cottonseed antigen from the gall bladder into the circulation.

The absorption time of the cottonseed from the gall bladder in this animal was six minutes.

Experiment 2.—In the above experiment, in which the common duct was clamped, one could not exclude the possibility that some of the antigen which had been injected into the lumen of the gall bladder may have escaped into the cystic duct and common duct. If this had occurred, some of the antigen could have been absorbed through the liver. It was necessary, therefore, to eliminate this possibility by clamping the cystic duct. The experiment was performed as follows:

A female monkey, weighing 2.25 kg., was used in this study.

Feb. 11, 1938: A cutaneous site on each thigh was sensitized to cottonseed with "P" serum as previously described.

Feb. 13, 1938. 10:55 A.M.: Narcosis was induced by the intravenous injection of 0.7 grain of nembutal.

11:00 A.M.: The abdomen was opened. The biliary tract was exposed. The cystic duct was clamped high up close to the gall bladder.

11:22 A.M.: Two-tenths milliliter of bile was withdrawn from the gall bladder by syringe, and the needle was left in situ. Five-tenths milliliter of cottonseed "milk" was then injected into the lumen of the gall bladder, through the same needle.

11:27 A.M.: Wheal formation started to develop at both sensitized cutaneous sites marking the entrance of cottonseed protein into the circulation.

The absorption time from the gall bladder in this animal was five minutes.

The above experiments have shown that, with the common duct or the cystic duct clamped, absorption of detectable traces of unaltered protein from the gall bladder takes place in the *Rhesus* monkey. Since the absorption phenomena demonstrated in the *Rhesus* monkey have closely simulated those observed in humans in every detail,[5] it is reasonable to assume that, under experimental conditions similar to those

described above, the absorption of unaltered protein could also be demonstrated from the gall bladder in humans.

It is of interest to note that the presence of bile in the gall bladder did not prevent the absorption of protein.

CONCLUSION

The absorption of detectable traces of unaltered protein has been shown to occur from the gall bladder in the *Rhesus* monkey.

REFERENCES

1. Harten, M., Gray, I., Livingston, S., and Walzer, M.: Studies in Absorption of Undigested Protein. IX. Absorption From the Stomach and Esophagus, J. Lab. & Clin. Med. (To be published.)
2. Gray, I., and Walzer, M.: Studies in Absorption of Undigested Proteins in Human Beings. VII. The Absorption of Unaltered Protein Introduced by Duodenal Tube Into the Abnormal Gastro-Intestinal Tract, Am. J. Digest. Dis. **5:** 345, 1938.
3. Gray, I., Harten, M., and Walzer, M.: Studies in Mucous Membrane Hypersensitiveness. IV. The Allergic Reaction in the Passively Sensitized Mucous Membranes of the Ileum and Colon in Humans, Ann. Int. Med. **13:** 11, 1940.
4. Gray, I., and Walzer, M.: Studies in Absorption of Undigested Protein in Human Beings. VII. Absorption From the Rectum and a Comparative Study of Absorption Following Oral, Duodenal, and Rectal Administrations, J. ALLERGY **11:** 3, 1940.
5. Straus, H. W., and Walzer, M.: Studies in Experimental Hypersensitiveness in the Rhesus Monkey. V. The Technic for Demonstrating the Absorption of Undigested Protein, J. ALLERGY **9:** 2, 1938.
6. Coca, A. F., and Grove, E.: Studies in Hypersensitiveness. XIII. A Study of the Atopic Reagins, J. Immunol. **10:** 445, 1925.

STUDIES ON BLOOD HISTAMINE IN CASES OF ALLERGY

III. The Effect of the Administration of Histaminase on the Symptoms and the Histamine Content of the Blood in Patients With Allergy

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WITHIN the past several years numerous investigators have endeavored to treat patients with allergic disease by the administration of histaminase, the specific enzyme capable of inactivating histamine. The rationale of such treatment rests on two theories, (1) the symptoms of anaphylactic shock are due to a liberation of histamine and (2) allergic disease in man is the counterpart of anaphylaxis. If such were the case, it would be reasonable to assume that the symptoms of allergy are also due to the release of histamine.

The release of histamine in anaphylactic shock has been amply demonstrated in the dog[10] and guinea pig.[7] However, in other species this does not obtain, for it was shown that a decrease in the histamine content of the blood occurs during anaphylactic shock in the horse and calf.[8] It was also demonstrated that an acute decrease in the histamine content of the blood of the rabbit occurs during anaphylactic shock[24] and that there is a moderate decrease in the histamine content of the tissues at the same time.[20] Thus, when the symptoms of anaphylaxis are manifest, most of the histamine has disappeared or has been inactivated in this species.

There has been much indirect evidence in favor of a release of histamine in certain types of allergic phenomena such as dermatographia[12, 17] and cold allergy,[11, 25] and it has been shown recently by direct estimation that a transient increase in the blood histamine may occur in patients with dermatographia or physical allergy when symptoms are induced.[18, 19] Although the blood histamine of patients with other types of allergic disease may fluctuate markedly, it has not been possible to demonstrate any correlation between these changes in the histamine level of the blood and the presence of symptoms, with the possible exception of angioneurotic edema where the histamine content of the blood is usually decreased.[18, 19] It will be seen then that there is still much to be explained about the role of histamine in anaphylactic shock in certain animal species and, especially, in allergic disease in man.

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During the course of observations on patients with allergic disease, it was decided to study the effect of the administration of histaminase both orally and intramuscularly on the symptoms and the histamine content of the blood.

METHODS

(a) *Type and Dosage of Histaminase.*—The preparations of histaminase* used are put up in the tablet form for oral use and as a dry, sterile, water-soluble powder in ampoules for intramuscular injection. The dose is measured in units, 1 unit being that amount of histaminase powder which is capable of inactivating 1 mg. of histamine when incubated with it for twenty-four hours at 38° C. The histaminase preparations were tested in this manner frequently for potency, and only active preparations were used. With the oral form, from 30 to 150 units were administered daily in three to four equally divided doses. Intramuscularly, histaminase was usually given in daily doses of from 1 to 8 units.

(b) *Histamine Determinations.*—As in the previous studies, histamine determinations were carried out by the Code[6] modification of the Barsoum and Gaddum method.[3] All results are expressed as gamma per 100 c.c. of whole blood as histamine base (gamma or γ = microgram or 0.001 mg.). It has been found from previous observations that the histamine content of the blood of normal persons is 4.0 γ/100 c.c. with variations of from 2.0 to 7.5 γ/100 c.c.[22]

RESULTS

Thirty-six patients were treated. The blood histamine was determined before and at intervals during and after the administration of the histaminase. Usually an interval of one week or ten days was allowed to elapse, and treatment was again resumed. During this period blood histamine levels were determined when possible. The results obtained over varying periods of time are given for ten patients (Table I).

It will be noted that there was little variation in histamine values in the periods of treatment as compared to those when no treatment was being given. Thus, in Case 121 the blood histamine remained at 6.0 γ/100 c.c. for most of the time. The last time the blood histamine was determined, it was 10.0 γ/100 c.c. There was no change in the symptoms. It has been previously noted that even greater fluctuations in the blood histamine may occur in patients with allergy without any alteration in symptoms.[18, 19] This is better illustrated in Case 60, where marked increases and decreases were observed during an acute attack

*The preparations of histaminase were supplied by the Winthrop Chemical Company, Montreal, through the courtesy of Dr. H. Cave.

TABLE I

THE EFFECT OF THE ADMINISTRATION OF HISTAMINASE ON THE SYMPTOMS AND HISTAMINE CONTENT OF THE BLOOD OF ALLERGIC PATIENTS

CASE NO.	SEX	AGE (YEARS)	DIAGNOSIS	DATE	TREATMENT	SYMPTOMS	HISTAMINE CONTENT OF THE BLOOD (γ/100 C.C.)
63	M	24	Urticaria	7/12/39	30 units orally started	Moderate	7.0
				7/16/39	45 units orally	No change	8.0
				7/21/39	Discontinued	No change	7.0
133	F	38	Urticaria	2/21/40	45 units orally started	Daily attacks moderate	3.5
				3/13/40	Increased to 75 units	No change	3.8
				3/20/40	Discontinued	No change	
				4/16/40	75 units orally started, and 2 units I.M.	Moderate	4.0
				4/22/40	Discontinued	Severe reaction in buttocks from the injections	0.5
121	F	32	Urticaria	2/14/40	45 units orally started	Moderate symptoms	6.0
				2/21/40	Same	Slight improvement	6.0
				2/28/40	Discontinued	Little change	6.0
				3/13/40	None	No change	
				4/ 2/40	90 units daily, orally and 2 units I.M. every second day started	No change	6.0
				4/ 8/40	Same as above	Feels better	6.0
				4/12/40	Discontinued	No relief	10.0
97	F	19	Chronic urticaria	1/ 8/40	None	Moderate daily attacks	7.0
				1/10/40	45 units orally started	Same as above	7.0
				1/15/40	Same as above	Same as above	
				1/25/40	Increased to 75 units	Severe attacks	10.0
				1/26/40	Discontinued	Severe attacks	6.0
				3/14/40	45 units daily orally and 2 units I.M. daily, started	Severe attacks	6.0
				3/20/40	Same as above	Severe attacks	6.0
				3/23/40	Discontinued	Severe attacks	6.0
				3/27/40	75 units daily orally and 4 units I.M. started	Moderate	5.5
				1/30/40	Discontinued	Moderate	
				4/15/40	One more attempt made unsuccessfully		

TABLE I (CONT'D)

CASE NO.	SEX	AGE (YEARS)	DIAGNOSIS	DATE	TREATMENT	SYMPTOMS	HISTAMINE CONTENT OF THE BLOOD (γ/100 C.C.)
189	M	10	Urticaria	4/ 8/40	45 units orally started	Moderate attacks	4.0
				4/12/40	Discontinued	Free of symptoms	5.0
				4/21/40	None	Severe attack	
				4/24/40	45 units orally started	No change	4.0
				4/27/40	Increased to 75 units	Severe	
				4/30/40	Same as above	Severe	4.0
				4/ 5/40	Discontinued	No change	4.0
				4/13/40	None	Free of symptoms	
82	F	49	Asthma	11/15/39	None	Acute attack	1.0
				11/16/39	None	Free	1.0
				12/ 5/39	45 units orally started	Acute attack	5.0
				12/ 6/39	Same	Improved	0.5
				12/11/39	Discontinued	Free of asthma	1.0
153	M	21	Colitis	3/13/40	None	8-10 B.M. daily	9.0
				5/23/40	None	5-6 B.M. daily	14.0
				5/31/40	75 units orally started	7-8 B.M. daily	10.5
				6/15/40	Increased to 150 units	12-14 B.M. daily with some blood	14.0
				6/20/40	Same as above	No change	9.0
				6/21/40	150 units orally and 8 units I.M.	No change	6.5
				6/28/40	Discontinued	No change	8.0
				7/ 2/40	None	Feeling better; 7-8 B.M. daily	
				7/ 3/40	150 units orally and 4 units I.M.	Symptoms worse	
				7/ 8/40	Discontinued	No change	7.0
197	F	36	Urticaria, headaches	4/18/40	45 units orally started	Moderate	2.8
				4/23/40	Increased to 75 units	No change	
				4/26/40	Same as above	Occasional headache; free of urticaria	2.0
				4/29/40	Discontinued	Severe headaches, urticaria	2.8

TABLE I (CONT'D)

CASE NO.	SEX	AGE (YEARS)	DIAGNOSIS	DATE	TREATMENT	SYMPTOMS	HISTAMINE CONTENT OF THE BLOOD (γ/100 C.C.)
78	F	14	Vasomotor rhinitis	4/27/40	45 units orally	Moderate	3.5
				5/13/40	Discontinued	No change	2.5
				5/16/40	45 units orally started	Moderate	
				5/23/40	Discontinued	Severe	10.0
				5/29/40	45 units started	Mild	5.5
				6/ 5/40	Same	No change	3.0
				6/ 8/40	Increased to 90 units	Severe	5.5
				6/13/40	Discontinued	Mild	3.0
				6/19/40	None	No change	3.0
				7/11/40	90 units orally started	Severe	3.0
				7/20/40	Discontinued	No change	
				9/11/40		Moderate	
				9/18/40	8 units daily I.M. started	Severe	
				9/24/40	Discontinued; started 80 units daily orally	Worse	5.8
				10/ 1/40	Discontinued	Slight improvement	10.0
60	M	18	Asthma, dermatitis	5/13/39	None	Severe dermatitis	12.0
				6/ 2/39	None	Skin improving; asthma beginning	14.0
				6/ 5/39	9 units daily I.M. started	Acute asthma	18.0
				6/11/39	Discontinued	Marked relief of asthma	16.0
				6/12/39	None	Free of asthma; skin clearing	9.0
				6/14/39	None	Free of asthma	
				6/15/39		Beginning asthma	
				6/16/39	30 units orally started	Free of asthma; dermatitis increasing	
				6/19/39	Discontinued		
				6/26/39	None	Free of asthma	13.5
				7/ 6/39	None	Severe asthma	
				7/ 7/39	None	Severe asthma	
				7/ 8/39	None	Free of symptoms	
				7/ 9/39	45 units daily	Beginning asthma	
				7/10/39	Same as before	Free of symptoms	

TABLE I (CONT'D)

CASE NO.	SEX	AGE (YEARS)	DIAGNOSIS	DATE	TREATMENT	SYMPTOMS	HISTAMINE CONTENT OF THE BLOOD (γ/100 C.C.)
				7/19/39	Same as before	Claims he can free himself within 3 days by taking histaminase	
				7/31/39	None	Persistent dermatitis; free of asthma	12.0
				10/12/40	45 units daily	Severe asthma	12.0
				10/19/40	Same as before	No change	14.0
				10/28/40	120 units daily	Moderate asthma	14.0
				11/ 6/40	Same	Asthma becoming more severe	12.0
				11/ 9/40	Same	Severe asthma	
				11/16/40	Discontinued	Severe asthma	14.0
				2/ 3/41	120 units daily started	Moderate asthma	22.0
				2/ 5/41	120 units daily	No change	22.0

The oral histaminase was given in three or four divided daily doses. The intramuscular injections were also given in one or more daily injections unless otherwise indicated.

B.M., Bowel movements; I.M., intramuscularly.

of asthma and at other times when there were relatively no changes in the symptoms. Thus, in July, 1939, there was a marked increase in the histamine content of the blood which coincided with an attack of asthma. At that time this patient was given histaminase intramuscularly (3 units t.i.d.). The symptoms abated, and the histamine content of the blood decreased. However, on many subsequent occasions administration of histaminase by mouth and by intramuscular injection did not affect the symptoms or the histamine content of the blood. Furthermore, the fluctuations in the blood histamine did not always coincide with attacks of asthma or dermatitis.

With regard to the intramuscular use of histaminase, it was found that, of seven patients so treated, four developed mild to severe reactions characterized by marked swelling, heat, and tenderness of the injected area. It was found as previously reported that, with the development of such areas, there was a marked decrease in the blood histamine level similar to the changes occurring in angioneurotic edema.[18, 19] This is shown in Table I, Case 133.

Questionable results were obtained in only four of the thirty-six patients treated with histaminase. These cases are summarized in Table II.

TABLE II

RESULTS OF TREATMENT WITH HISTAMINASE ON 36 PATIENTS WITH ALLERGIC DISEASE

	DIAGNOSIS	NO.	IMPROVED	NO CHANGE
A. Oral Administration				
	Urticaria	15	2	13
	Asthma	11	1	10
	Vasomotor rhinitis	5	1	4
	Angioneurotic edema	3	0	3
	Eczema	1	0	1
	Total	36	4	32
*B. Intramuscular Injection**		7	0	7

*Local reactions to the injections in 4 cases.

DISCUSSION

The destruction of histamine takes place in the body by several mechanisms, one of which depends upon the enzyme histaminase. For example, it was shown that tissues containing no histaminase were much more active in the destruction of histamine in vivo in the rat than were those tissues which contained the enzyme.[21]

Even if histaminase were the main factor in the destruction of histamine, it is difficult to understand how the oral administration of this substance could be effective for, as Best and McHenry[5] have again pointed out, histaminase is inactivated by hydrochloric acid and would therefore be rendered inert if introduced into the stomach. They noted further that it is inactivated by pepsin in acid solution and by trypsin in alkaline solution. Recently Roth and Horton[25] have claimed that the oral administration of histaminase to patients could effectively inhibit the liberation of free hydrochloric acid in the stomach following the subcutaneous injection of histamine. This work was repeated by Atkinson, Ivy, and Bass,[1] who were unable to confirm these results. They were unable to alter the gastric secretion stimulated by histamine in patients or in dogs when they gave potent preparations of histaminase either orally or intramuscularly. Furthermore, the original contention of Karady and Browne[13] that the intravenous administration of histaminase could inhibit histamine shock in normal guinea pigs, or, if injected prior to the shock dose of antigen in sensitized guinea pigs, could inhibit anaphylaxis, has not been confirmed by recent observers.[5, 9, 15, 23, 26]

It is difficult to assess the value of the administration of histaminase in patients with allergy on symptoms alone. Reports in favor of the efficacy of histaminase are based mostly on this criterion. For example, Laymon and Cumming[16] treated seventeen cases of urticaria, claiming good results in 60 per cent after the administration of his-

taminase for ten days. Baker[2] obtained good results in two cases of cold allergy by the combined treatment with desensitization to cold and the oral administration of histaminase. Roth and Horton[25] have also claimed good results in cases of allergy by the oral administration of histaminase. Keeney[14] has, however, shown that the fallacy of accepting a decrease in symptoms as the sole criterion of efficacy of histaminase administration is a real one, for, when his patients with hay fever improved on histaminase therapy, there was also a coincident decrease in pollen counts of the atmosphere which in his judgment accounted for the decrease in symptoms.

It is well known that most of the allergic diseases are characterized by spontaneous remissions of the symptoms. If the histaminase is given before such a remission, the results are apt to be misinterpreted, and this is shown in the results obtained on several of the patients described in the present report.

The results of the present communication would indicate that the administration of histaminase either orally or by intramuscular injection to patients with allergic disease has no effect on either the symptoms or the histamine content of the blood. It is possible that highly active preparations may have a demonstrable effect but, as yet, these are not available.

CONCLUSIONS

1. The histamine content of the blood and symptoms of thirty-six patients with allergic disease have been observed during the administration of histaminase both orally and intramuscularly.

2. No effect on the histamine content of the blood was noted in all patients following or during the administration of the histaminase. In four cases questionable improvement in symptoms was observed.

3. In four of the seven patients treated by intramuscular injection of histaminase, reactions consisting of swelling, redness, and tenderness at the site of injection were observed.

REFERENCES

1. Atkinson, A. J., Ivy, A. C., and Bass, V.: Am. J. Physiol. **132:** 51, 1941.
2. Baker, T. W.: J. A. M. A. **114:** 1059, 1940.
3. Barsoum, G. S., and Gaddum, J. H.: J. Physiol. **85:** 1, 1935.
4. Best, C. H., and McHenry, E. W.: J. Physiol. **70:** 349, 1930.
5. Best, C. H., and McHenry, E. W.: Canad. M. A. J. **43:** 163, 1940.
6. Code, C. F.: J. Physiol. **89:** 257, 1937.
7. Code, C. F.: Am. J. Physiol. **127:** 78, 1939.
8. Code, C. F., and Hester, H. H.: Am. J. Physiol. **127:** 71, 1939.
9. Corper, H. J., and Cohn, M. L.: J. A. M. A. **115:** 30, 1940.
10. Dragstedt, C. A., and Gebauer-Fuelnegg, E.: Am. J. Physiol. **102:** 512, 1932.
11. Horton, B. T., Brown, G. E., and Roth, G. M.: J. A. M. A. **107:** 1263, 1936.
12. Kalk, H.: Klin. Wchnschr. **8:** 64, 1929.
13. Karady, S., and Browne, J. S. L.: J. Immunol. **37:** 463, 1939.

14. Keeney, E. L.: J. A. M. A. **114**: 2448, 1940.
15. Knoll, A. F.: Proc. Soc. Exper. Biol. & Med. **45**: 606, 1940.
16. Laymon, C. W., and Cumming, H. A.: J. Invest. Dermat. **2**: 301, 1939.
17. Lewis, T.: The Blood Vessels of the Human Skin and Their Responses, London, 1927, Shaw & Sons, Ltd.
18. Rose, B.: J. ALLERGY **12**: 327, 1941.
19. Rose, B.: J. Clin. Investigation (in press).
20. Rose, B.: Am. J. Physiol. **129**: 450, 1940.
21. Rose, B., and Browne, J. S. L.: Am. J. Physiol. **124**: 412, 1938.
22. Rose, B., and Browne, J. S. L.: Proc. Soc. Exper. Biol. & Med. **44**: 182, 1940.
23. Rose, B., and Browne, J. S. L.: J. Immunol (in press).
24. Rose, B., and Weil, P.: Proc. Soc. Exper. Biol. & Med. **42**: 494, 1939.
25. Roth, G. M., and Horton, B. T.: Bull. New York Acad. Med. **16**: 570, 1940.
26. Youngner, J. S., Freedman, S. I., and Nungester, W. J.: Proc. Soc. Exper. Biol. & Med. **46**: 187, 1941.

THE BLOOD HISTAMINE LEVEL IN ASTHMA AND IN EOSINOPHILIA

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THERE is good reason to suspect that the level of blood histamine might be elevated in patients with bronchial asthma, particularly when studied during an attack. Dale and Laidlaw[1] were the first to call attention to the similarity between the symptoms of histamine shock and anaphylactic shock in various species of animals. Weiss, Robb, and Blumgart[2] have shown that asthmatic patients react to histamine more markedly than normal patients and that the intravenous injection of histamine to patients with a history of bronchial asthma produced unmistakable signs of bronchospasm. It thus seemed desirable to study the blood histamine levels during acute attacks of asthma in comparison with the values between attacks and with the values found in a group of nonasthmatic patients. Concurrent white blood cell and eosinophile cell counts were made in addition to the histamine determinations, since Code[3, 4] had shown that from 70 to 100 per cent of the histamine-like substance extractable from the whole blood of man and various animals was contained in the white blood cell layer and particularly in the eosinophiles. Our study also includes blood histamine observations on a few patients with a high degree of blood eosinophilia.

The technique of blood histamine determination used was that described by Barsoum and Gaddum[5] in 1935 and modified by Code[6] in 1937. The principles are as follows: Because of the stability of histamine, other vasodilator substances may be removed by precipitation of whole blood with trichloracetic acid and hydrolysis of the filtrate by boiling with HCl. After the excess acid has been removed by repeated evaporation in the presence of alcohol, the dried residue is extracted with water, neutralized, and made up to the volume of the original sample. Pharmacologic assays are then made upon a guinea pig intestinal strip in comparison with a standard solution of histamine phosphate. The smallest amount of the unknown solution which will give a distinctly stronger contraction of the intestinal strip than an amount of the standard solution of histamine is found; then the largest amount of the unknown giving a distinctly weaker contraction is determined. The middle value is taken to correspond with the standard. All determinations are made in duplicate. The values are averaged and recorded in terms of gamma (micrograms) histamine base per cubic centimeter of whole blood.

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Accuracy in the use of the method was studied by attempting the recovery of histamine added to normal untreated blood. From 70 to 78 per cent of added histamine could be recovered when dealing with quantities between 0.66 and 1.0 gamma per cubic centimeter. Although Code reported approximately 90 per cent recovery, he added histamine to blood from which the naturally occurring histamine had been removed by perfusion through a dog's kidney.

TABLE I

BLOOD HISTAMINE EQUIVALENTS IN 20 NORMAL PATIENTS

PATIENT	W.B.C. PER C.MM. OF BLOOD	EOSINOPHILES PER C.MM. OF BLOOD		HISTAMINE BASE (GAMMA PER C.C. OF WHOLE BLOOD)
		NO.	%	
J. C.	5,800	58	1	0.064
E. P.	6,700	201	3	0.053
H. R.	8,250	165	2	0.071
M. C.	8,400	168	2	0.039
J. P.	9,200	276	3	0.042
G. M.	7,450	149	2	0.068
E. W.	10,200	510	5	0.019
M. M.	7,600	0	0	0.034
A. P.	11,150	111	1	0.027
F. D.	10,100	202	2	0.034
H. H.	8,550	427	5	0.033
K. B.	5,650	113	2	0.032
J. B.	7,400	222	3	0.047
M. F.	8,600	86	1	0.068
B. C.	6,400	128	2	0.030
A. F.	9,250	185	2	0.056
G. S.	8,700	348	4	0.070
F. R.	8,000	160	2	0.051
B. B.	7,300	292	4	0.034
T. R. (a)	9,100	273	3	0.039
(b)	6,850	137	2	0.033
(c)	7,400	222	3	0.038
Av.	6,670	201	2.5	0.045

Histamine determinations and white blood cell counts were made in a series of twenty normal nonallergic adults who had visited the hospital because of minor surgical complaints not associated with infections. The histamine equivalents ranged from 0.019 to 0.071, averaging 0.045 gamma per cubic centimeter. These values are in close agreement with a series of fifty control observations reported by Rose and Browne[7] in which Code's modification of Barsoum and Gaddum's method was also used. They reported a variation from 0.025 to 0.081 and an average of 0.04 gamma per cubic centimeter.

THE HISTAMINE CONTENT OF THE BLOOD IN PATIENTS WITH BRONCHIAL ASTHMA

The patients in this group had typical histories and physical findings of bronchial asthma. They were not only subject to attacks of asthma

of great severity but between attacks were comparatively free of asthmatic symptoms. Table II records the blood counts and histamine determinations in nine patients during attacks of asthma and in six of these during intervals between attacks. The blood samples taken during paroxysms of asthma were drawn at the height of symptoms after at least three hours had elapsed since the administration of any medication. The determinations between seizures were made when the patients were comfortable and the lungs were free of râles and when at least twelve hours had elapsed since the last administration of epinephrine or ephedrine.

TABLE II

BLOOD HISTAMINE EQUIVALENTS IN BRONCHIAL ASTHMA

	DURING ATTACKS OF ASTHMA					BETWEEN ATTACKS OF ASTHMA			
PATIENT	SEVERITY OF ASTHMA	W.B.C. PER C.MM. OF BLOOD	EOSINOPHILES PER C.MM. OF BLOOD		HISTAMINE BASE (GAMMA PER C.C. OF BLOOD)	W.B.C. PER C.MM. OF BLOOD	EOSINOPHILES PER C.MM. OF BLOOD		HISTAMINE BASE (GAMMA PER C.C. OF BLOOD)
			NO.	%			NO.	%	
M. O.	Very severe	8,200	0		0.160	7,300	73	1	0.083
J. S.	Very severe	9,100	364	4	0.098	8,400	252	3	0.057
M. C.	Mod. severe	7,850	706	9	0.097	8,200	490	6	0.037
		9,100	819	9	0.078				
H. H.	Severe	7,400	444	6	0.080	8,100	405	5	0.037
						9,700	338	4	0.054
M. M.	Mod. severe	8,400	504	6	0.088	9,100	1,183	13	0.069
M. G.	Mod. severe	7,800	312	4	0.048	6,500	195	3	0.035
J. M.	Severe	9,350	1,122	12	0.057				
	Severe	9,100	1,365	15	0.053				
P. G.	Very severe	6,450	258	4	0.107				
M. R.	Severe	8,700	522	6	0.077				
Av.					0.088				0.054

Seven of the nine patients studied had higher histamine levels during attacks of asthma than were observed in any control cases; the greatest elevation (0.160 gamma per cubic centimeter) occurred in case M. O., who had the most violent attack of any of the patients observed and who died during an attack of asthma four months after this determination. In six cases determinations were made both during and between attacks of asthma; each patient had a higher histamine equivalent during acute symptoms than when symptom-free, although in no instance was there more than a two-fold increase.

The degree of elevation of the blood histamine level during attacks of asthma in this group agrees with the findings of Haworth and Macdonald,[8] who reported elevated blood histamine values during the presence of asthmatic symptoms (average of 0.06) in sixty-five card-

room workers as compared with those in University students (average of 0.04) or in patients hospitalized for bronchitis (average of 0.03 gamma per cubic centimeter).

Observations on the blood histamine levels in bronchial asthma were first made by Cerqua[9] in 1936. He studied eight cases of asthma and found that the histamine equivalents during attacks ranged from 0.180 to 0.220, reached a maximum of 0.210 to 0.300 at the end of an attack, and usually returned to the normal of 0.050 to 0.070 gamma per cubic centimeter after ten hours. In some patients the histamine values between paroxysms remained slightly greater than his maximal normal figures; they ranged from 0.070 to 0.100 gamma per cubic centimeter.

Parrot[10] in 1938 studied the blood in four cases of asthma and confirmed the finding of Cerqua that the blood histamine equivalents during asthmatic crises were elevated from three to four times normal values.

All of these workers used Barsoum and Gaddum's method of analysis. When these blood histamine values in bronchial asthma are reviewed in the light of Code's improvement in technique, it is conceivable that a portion of the histamine content of these extracts may have been lost in the process of analysis.

In summary, the results in the present study revealed a moderate increase in the blood histamine content during acute asthmatic symptoms compared with the interval between attacks (an average of 0.088 as against 0.054 gamma histamine base per cubic centimeter of whole blood) and as compared with the blood levels found in normal patients (average of 0.045 gamma histamine base per cubic centimeter of whole blood). This variation is less marked than is reported by earlier investigators although these workers employed a method (Barsoum and Gaddum) reported to be slightly more destructive of histamine than was the case of the technique used in our study (Code's modification).

More recently, another Italian, Capuani[11] in 1939 reported a two-fold increase in the blood histamine in asthmatic patients during attacks as compared with nonasthmatic persons. His studies were based upon thirty-five cases of asthma and twelve controls which consisted of nonasthmatic tuberculous patients. The exact levels of blood histamine reported by him cannot be adequately compared with those found by other workers since he used an entirely different method of analysis. He depended upon removing the histamine present in a blood sample by absorption upon permutit and estimating the amount by colorimetry based upon Pauly's diazo reaction. By this procedure he found normal blood values approximately twenty times greater than those reported by the older methods. His technique is more simple than the complicated and technically difficult method of Barsoum and Gaddum as modified by Code, but whether it is as accurate remains to be determined.

THE HISTAMINE CONTENT OF THE BLOOD IN PATIENTS WITH A HIGH DEGREE OF BLOOD EOSINOPHILIA

Code's suggestion that the eosinophile cell may be closely associated with the histamine in the blood was based in part upon Macdonald's findings (as reported by Code) of an elevated blood histamine content in four cases of eosinophilia. One case associated with parasitosis had 650 eosinophiles per cubic millimeter of blood and a histamine equivalent of 1.00 gamma per cubic centimeter of blood. Another case of questionable parasitic etiology had upon three determinations an eosinophilia between 1,850 and 3,000 cells per cubic millimeter and a histamine level between 0.44 and 0.51 gamma per cubic centimeter. The two remaining cases, one of splenic anemia and one of unknown etiology, had between 960 and 1,230 eosinophiles per cubic millimeter with histamine values between 0.14 and 0.19 gamma per cubic centimeter.

TABLE III

BLOOD HISTAMINE EQUIVALENTS IN PATIENTS WITH EOSINOPHILIA

PATIENT	DIAGNOSIS	W.B.C. PER C.MM. OF BLOOD	EOSINOPHILES PER C.MM. OF BLOOD		HISTAMINE BASE (GAMMA PER C.C. OF BLOOD)
			NO.	%	
R. K.	Periarteritis nodosa and bronchial asthma	9,800	5,390	55	0.019
		14,000	7,000	50	0.025
		23,400	15,910	68	0.027
M. L.	Periarteritis nodosa and bronchial asthma	27,600	16,280	59	0.057
B. P.	Periarteritis nodosa and bronchial asthma	18,400	6,800	37	0.038
E. S.*	Periarteritis nodosa and bronchial asthma	27,200	12,240	45	0.105
H. F.	Bronchial asthma and chronic bronchitis	8,900	1,510	17	0.121
B. G.*	Bronchial asthma	22,200	5,770	26	0.074
G. D.*	Arsphenamine dermatitis	18,500	9,060	49	0.035
A. R.	Atrophic arthritis	8,700	1,910	22	0.044

*These cases were studied through the courtesy of the staff of the Lahey Clinic.

It seemed desirable, therefore, to determine the blood histamine equivalents in additional cases of eosinophilia. Eight such patients were studied, and, as shown in Table III, the eosinophilia ran to high percentages and in no instance was the etiology associated with the presence of a parasite. In four cases the marked eosinophilia was recognized as part of the clinical picture of periarteritis nodosa as described by Rackemann and Greene.[12] Two patients had asthma apparently without periarteritis; one had an extensive dermatitis due to arsphenamine; and one patient with atrophic arthritis was found to have a high blood eosinophilia. In all except one case the eosinophilia was over 20 per cent, and in four patients the differential count of blood showed over 40 per cent of eosinophiles.

The study of these patients was most interesting. In only two cases were the histamine values increased over 0.08 gamma per cubic centimeter; these slight elevations occurred in patients having mild asthma at the time the blood samples were drawn. Of these two cases, one had the highest histamine content of any in this series (0.121 gamma per cubic centimeter) but had the lowest total number of eosinophiles (1,510 per cubic millimeter). It will also be noted that five of the eight cases had considerably higher eosinophile counts than any of Macdonald's cases but had histamine equivalents within normal limits.

From the study of these eight cases as well as from the eosinophile observations in the controls and the asthmatic patients, it seems that there is no constant relationship between the number of eosinophiles in the peripheral blood and the level of blood histamine.

SUMMARY

1. Blood histamine determinations using Code's modification of Barsoum and Gaddum's method were made on twenty normal patients having normal white blood cell and eosinophile counts. The values averaged 0.045 and ranged from 0.019 to 0.071 gamma (micrograms) histamine base per cubic centimeter of whole blood.

2. Simultaneous blood histamine and eosinophile determinations were made in nine patients with bronchial asthma. There was a suggestive increase above normal in the blood histamine equivalent during paroxysms of asthma compared with the interval between attacks.

3. Normal blood histamine equivalents were found in six patients having high degrees of blood eosinophilia. Slightly elevated blood histamine values were observed in two additional cases of eosinophilia; both patients were having mild asthma at the time the blood samples were taken.

4. There appeared to be no constant relationship between the number of eosinophiles in the peripheral blood and the level of the blood histamine equivalents.

We wish to thank Dr. Otto Krayer of the Pharmacology Department, Harvard Medical School, for his valuable suggestions and supervision during the course of this study.

REFERENCES

1. Dale, H. H., and Laidlaw, P. P.: The Physiological Action of B-Iminazolylethylamine, J. Physiol. **41**: 318, 1910.
2. Weiss, S., Robb, G. P., and Blumgart, H. L.: The Velocity of Blood Flow in Health and Disease as Measured by the Effect of Histamine on Minute Vessels, Am. Heart J. **4**: 664, 1929.
3. Code, C. F.: The Source in Blood of the Histamine-Like Constituent, J. Physiol. **90**: 349, 1937.
4. Code, C. F.: The Histamine-Like Activity of White Blood Cells, J. Physiol. **90**: 485, 1937.
5. Barsoum, G. S., and Gaddum, J. H.: The Pharmacological Estimation of Adenosine and Histamine in the Blood, J. Physiol. **85**: 1, 1935.

6. Code, C. F.: The Quantitative Estimation of Histamine in Blood, J. Physiol. **89:** 257, 1937.
7. Rose, B., and Browne, J. S. L.: Alterations in the Blood Histamine in Shock, Proc. Soc. Exper. Biol. & Med. **44:** 182, 1940.
8. Haworth, E., and Macdonald, A. D.: On Histamine in Cotton Dust and in Blood of Cotton Workers, J. Hygiene **37:** 234, 1937.
9. Cerqua, S.: L'Equivalente Istaminico nel Sangue in Diverse Condizioni, Minerva med. **1:** 542, 1936.
10. Parrot, J. L.: Les Manifestations de L' Anaphylaxie et les Substances Histaminiques, Paris, 1938, J. B. Baillière et Fils.
11. Capuani, G. F.: Istaminemia nell Asma Bronchiale, Riforma med. **55:** 1539, 1939.
12. Rackemann, F. M., and Greene, J. E.: Periarteritis Nodosa and Asthma, Tr. A. Am. Physicians **54:** 112, 1939.

COSEASONAL INTRACUTANEOUS TREATMENT OF HAY FEVER

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THE coseasonal intracutaneous method of treatment of hay fever was first reported about fifteen years ago by Duke,[1] Phillips[2] and Thommen.[3] Later, in 1932 and 1935, Anderson[4, 5] reported his observations on the use of this method of therapy. More recently a number of brief reports have appeared in the letters of the International Correspondence Club of Allergy.

Some observers have advocated the intracutaneous method only, beginning treatment shortly before or at the onset or during the hay fever season. Others recommended combined intracutaneous and subcutaneous injections given either at the same visit or starting with one or more intracutaneous injections followed by subcutaneous injections only.

Phillips[2, 6] reported his first observations in 1926 and submitted a further report in 1933 on a group of 322 patients treated over a period of six years. Before starting treatment the patients were quantitatively tested for the degree of sensitivity. The first dose was usually 0.02 c.c. of the 1:5,000 solution. Dosages were gradually increased until the local reaction reached the size of the patient's palm. When more than 0.17 c.c. was necessary, the dosage was divided and given in two sites. Relief was usually obtained by the end of seven days of intensive treatment. In the group of 322 patients satisfactory results were obtained in about 91 per cent. Twelve general reactions occurred in nine instances, or one to each of 625 injections.

Anderson[4, 5] also advocated the small dosage type of intracutaneous coseasonal treatment for hay fever. In a group of 350 patients, 299 never received a total of more than 0.10 c.c. of a 1:4,000 dilution, and forty-two never received more than a total of 0.10 c.c. of a 1:500 dilution. All treatments were given by the intracutaneous route at intervals of one day in severe cases and every ten to fourteen days in patients having mild symptoms. For the first injection Anderson recommended 0.01 c.c. of a 1:40,000 dilution. The greatest volume reached was 0.05 c.c. of a 1:4,000 dilution of any single or combined pollen mixture. In a group of fifty-five patients who had previously received the small dosage method of treatment, twenty-four required no further injections because of complete freedom from symptoms. Other patients required only occasional treatments in order to remain comfortable.

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Matzger[7] employed the same type of treatment recommended by Phillips.[2, 6] Small doses were given intracutaneously until the patient was symptom-free and continued either subcutaneously or intracutaneously as needed. Dutton[8] also recommended the intracutaneous method of treatment.

Harris[9] advocated daily injections until relief was obtained, then injections every two to three days. An initial dose of 0.01 c.c. of the 1:10,000 dilution was given intracutaneously. Subsequent injections were increased slowly until a local reaction was obtained. A maximum dose was rarely more than 0.10 or 0.20 c.c. of the 1:10,000 dilution.

Davidson[10] used a combination of intracutaneous and subcutaneous treatments. Treatment was started with the strongest dilution the patient could take both subcutaneously and intracutaneously. In most instances he noted that the patient could take a stronger solution intracutaneously than subcutaneously. Doses were given every day until relief was obtained, then every two or three days, twice a week or once a week.

Seyler[11] started treatment with an intracutaneous dose which gave a 25 mm. wheal in twenty-five mintes. He also gave subcutaneous injections with 0.05 c.c. of the dilution which caused an intracutaneous reaction of 16 mm. Intracutaneous doses were usually not increased. The subcutaneous doses were increased just enough to produce a sore arm. The intracutaneous injections were given every other day and the subcutaneous dose as often as the subsidence of the previous dose allowed. The intracutaneous injections were discontinued after seven to ten days and further treatment continued by the subcutaneous method.

Brown[12] also employed the combined subcutaneous and intracutaneous methods. He stated that less extract was effective by the subcutaneous than by the intracutaneous route. Without the intracutaneous reactions he found it difficult to judge the dosage. Occasionally better results were noted by the intracutaneous than by the subcutaneous method.

Barnes[13] advocated an initial dosage intracutaneously of 0.02 c.c. of the 1:2,500 dilution. Subsequent doses were given subcutaneously and gradually increased until relief was obtained. When this was reached, the intervals were increased to every two days or four to five days. The maximum dose was 60 to 80 units. Barnes had some failures by the preseasonal method, even in patients who had previously had good results by the intracutaneous method.

Schonwald[14] gave an initial dose of 0.10 c.c. of extract intracutaneously. He started with a concentration ten times stronger than the one which gave a one-plus reaction by the intracutaneous test. Intracutaneous injections were repeated until the skin reactions became weaker, i.e., usually after four or five days. Then subcutaneous treatment was started cautiously. In most cases he began with a dilution of 1:100,000, sometimes 1:1,000,000, and occasionally as strong as the 1:10 dilution.

Halpin[15] discarded the preseasonal method in favor of the coseasonal because of better results. He started with the intracutaneous injections and continued with the subcutaneous. The maximum dosage was rarely above 0.10 c.c. of the 1:1,000 dilution. The injections were given daily, twice a week, or even at five-day intervals. The intervals were determined by the amount of local reaction at the site of the previous injection as well as by the duration of relief afforded by the extract.

In the specific coseasonal method recommended by Wittich[16] for ragweed hay fever the initial dosage was determined on the basis of the concentration which gave a local reaction with the scratch test. When this concentration was 0.001 mg. of nitrogen per cubic centimeter, then 0.10 c.c. of the 1:10,000 dilution (10 units) was given as the initial dose subcutaneously. When 0.0001 mg. of nitrogen per cubic centimeter was the proper concentration, then 0.10 c.c. of the 1:100,000 extract (1 unit) was given. Dosages were gradually increased by 0.10 c.c. with each subsequent injection until 1.0 c.c. was given. Wittich found that the most satisfactory results were obtained when the maximum dosage did not go above 0.10 c.c. of the 1:1,000 dilution (100 units). Injections were given at intervals of two to three days, and about twelve to fourteen doses were required for the season.

After determination of the degree of skin sensitivity, Vaughan[17] used the subcutaneous method of treatment. Adequate relief was noted with not more than 80 to 100 Noon-Coca units as a maximum dose. The initial dose of 20 units was gradually increased according to the effect obtained. A 1:5,000 dilution of the extract was employed. When the optimum dosage was reached, the intervals between injections were increased.

In these reports it has been shown that satisfactory relief can be obtained by the coseasonal method of treatment whether given only by the intracutaneous, by the subcutaneous, or by both routes of administration. The degrees of relief obtained by the doses of extract employed indicate that small amounts are effective. It is probable that many of the poor results obtained by the preseasonal or perennial methods are the result of overdosage rather than underdosage. By the coseasonal method it is evident that optimum dosage can be determined without difficulty.

Among the various advocates of the intracutaneous method, some have recommended the titration of skin sensitivity in order to determine the initial dosage. Others simply state that the initial dosage is started with a certain amount of a given dilution regardless of the degree of sensitivity. Only a few observers have recommended that the dosage be determined on the basis of the size of the wheal produced. This, we believe, is the most satisfactory method of regulating the dosage, because the degree and duration of relief of symptoms are generally proportional to the size of the wheal as well as to the amount and strength of the dilution employed.

With patients who did not receive satisfactory results from preseasonal therapy after the season began, the intracutaneous method was used and the subcutaneous injections were discontinued. Intracutaneous treatment was also found to be most satisfactory with patients who had had incomplete preseasonal therapy. Whenever possible the coseasonal type of therapy should be preceded by a series of about ten injections either intracutaneously or subcutaneously before the onset of the season.

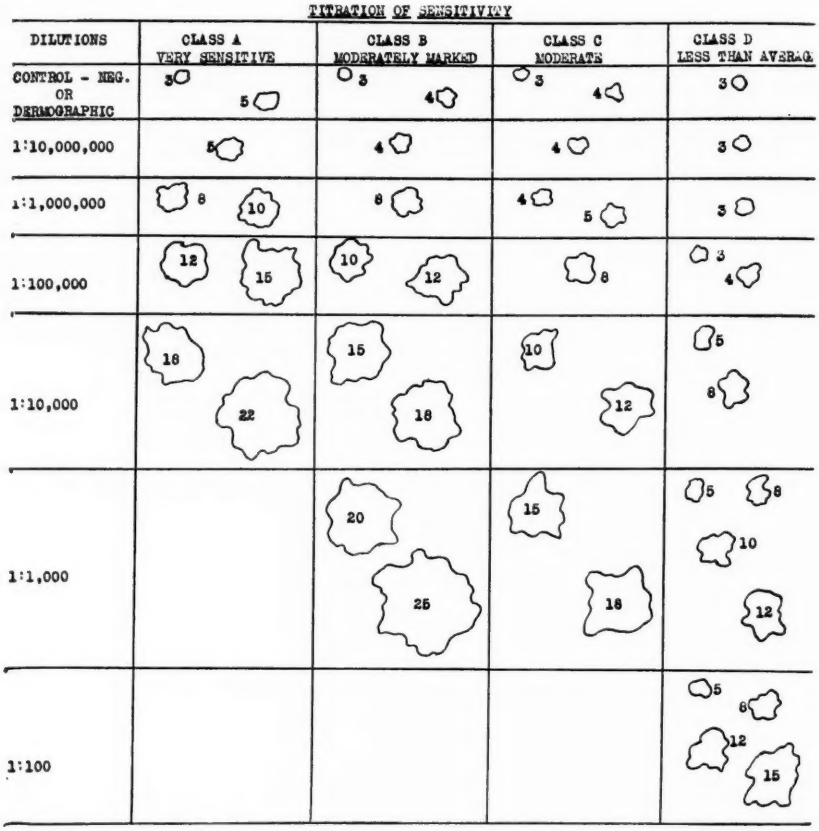


Fig. 1.—Treatment of hay fever caused by trees, grasses, and weeds. Always test first with 1:10,000,000, 1:1,000,000, and 1:100,000 dilutions; then add 1:10,000 and 1:1,000 dilutions according to sensitivity. (Diameters of wheals are given in millimeters.)

During the past six years we have employed the coseasonal method of treatment in practically all the tree and grass cases and in about one-half of the ragweed cases with most satisfactory results. Most of the patients had ragweed hay fever; many of these did not report for treatment until after the season had started. Some patients were treated by the intracutaneous method for six consecutive years.

Before the initial dosage is given, the skin sensitivity is determined by intracutaneous tests with various dilutions ranging from 1:10,000,000

to 1:1,000 (Fig. 1). Occasionally the 1:100,000,000 or the 1:100 dilutions are necessary. Initial testing is always made with the 1:10,000,000, 1:1,000,000, and the 1:100,000 dilutions using Coca solution as a control. If a positive whealing reaction as large as 15 mm. in diameter is noted to the 1:100,000 dilution, no further tests are made at the initial sitting (very marked sensitivity, Fig. 1). Within one or two days treatment should be started with 0.02 c.c. of the 1:100,000 dilution which usually produces a wheal about 15 to 18 mm. in diameter. Subsequent doses should be administered every two to five days or longer according to the degree of relief obtained. The amount of the injections should be increased by 0.01 c.c. as shown in Table I. The size of the

TABLE I

DOSES OF POLLEN EXTRACTS FOR THE INTRACUTANEOUS METHOD OF HAY FEVER TREATMENT

NOON-COCA UNITS	TOTAL MG. NITROGEN	WEIGHT PER VOLUME STANDARDIZATIONS		
		DILUTION	MG. N PER C.C.	EXTRACT (C.C.)
0.10	0.000001	1:100,000	0.0001	0.01
0.20	0.000002			0.02
0.30	0.000003			0.03
0.40	0.000004			0.04
0.50	0.000005			0.05
1.0	0.00001	1:10,000	0.001	0.01
2.0	0.00002			0.02
3.0	0.00003			0.03
4.0	0.00004			0.04
5.0	0.00005			0.05
10.0	0.0001	1:1,000	0.010	0.01
20.0	0.0002			0.02
30.0	0.0003			0.03
40.0	0.0004			0.04
50.0	0.0005			0.05
60.0	0.0006			0.06
70.0	0.0007			0.07
80.0	0.0008			0.08
90.0	0.0009			0.09
100.0	0.001			0.10

wheal increases with each subsequent dosage by about 1 to 2 mm. The average size wheal which produces satisfactory relief varies from 20 to 25 mm. in diameter. Occasionally it may be necessary to increase it to 30 mm. in diameter. In order to reach optimum dosage, as much as 0.05 c.c. of the 1:10,000 dilution may be necessary in cases of very marked sensitivity. After a few treatments at two- to three-day intervals it is often possible to increase the intervals from five to seven days in ragweed cases. In the tree and grass cases longer intervals are often possible because of the marked variations in pollen concentration. In some of the grass cases intervals of two weeks or longer are possible.

With each subsequent treatment there is a tendency to reduction of the skin reaction in proportion to the dosage injected as shown in the case reports.

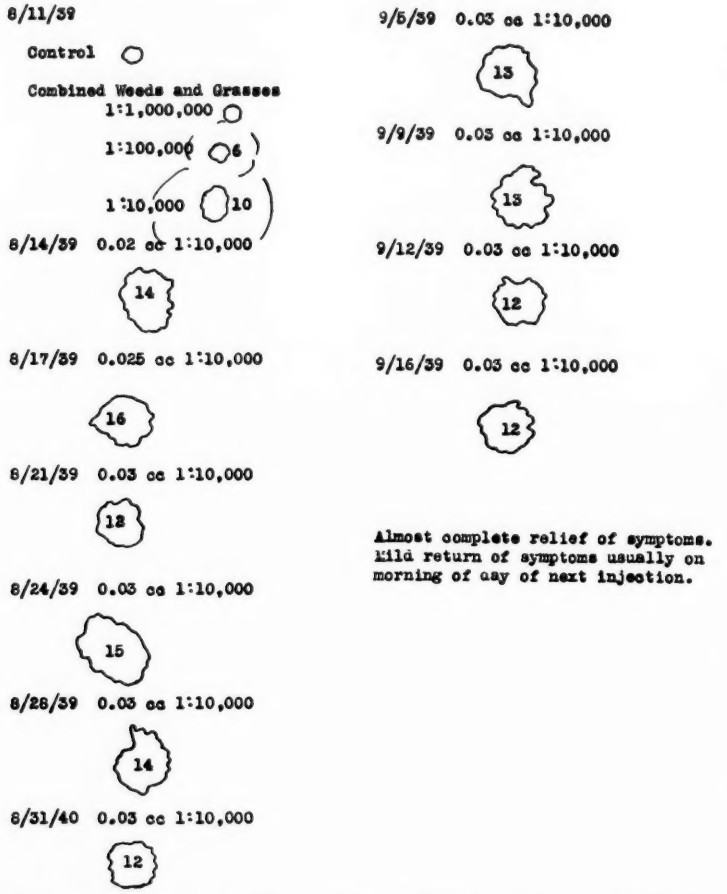


Fig. 2.—Case 1. B. P., aged 5 years. Ragweed hay fever. Positive scratch reactions to giant and short ragweed.

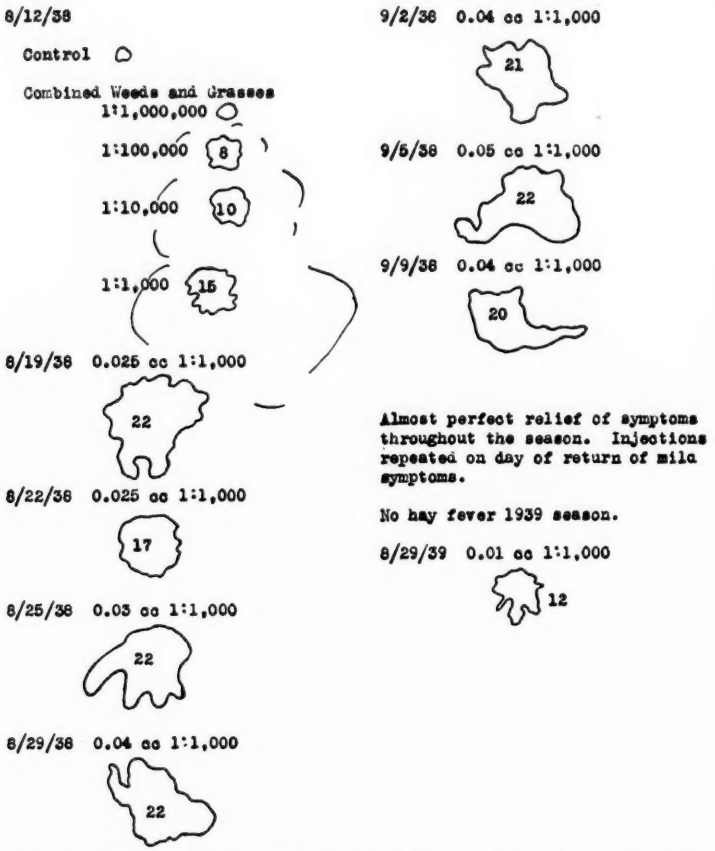


Fig. 3.—Case 2. L. E., aged 46 years. Ragweed hay fever. Fourth year of treatment.

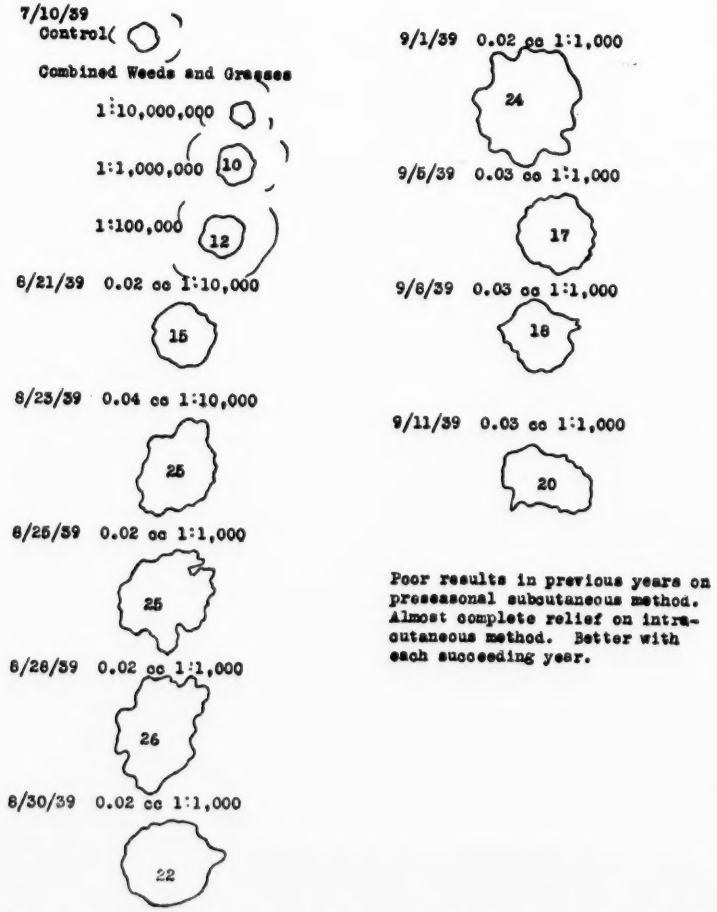


Fig. 4.—Case 3. J. R. L., aged 61 years. Ragweed hay fever. Fourth year of treatment.

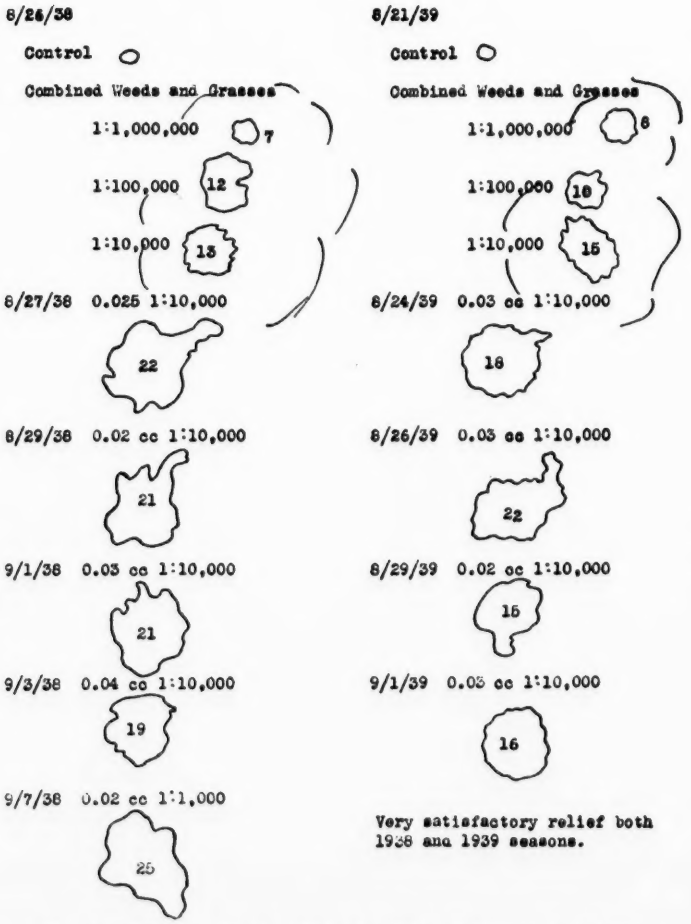
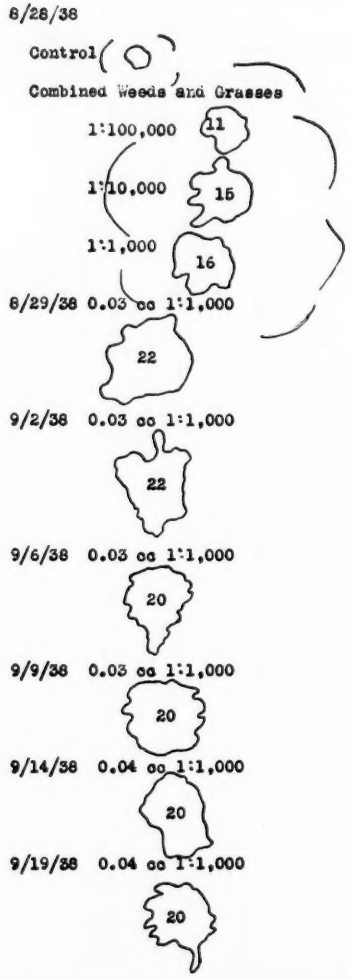


Fig. 5.—Case 4. B. H., aged 19 years. Ragweed hay fever.



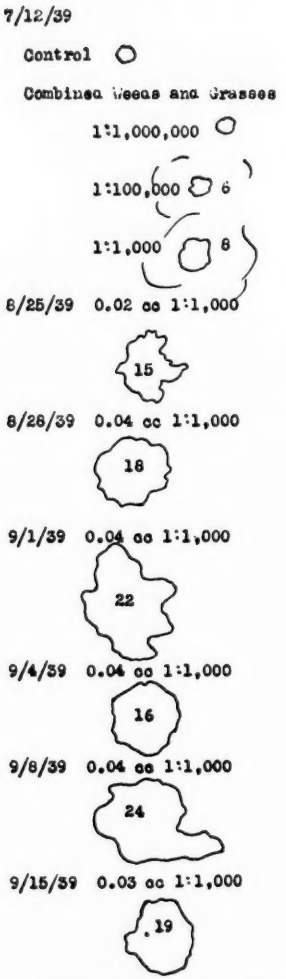


Fig. 6.—Case 5. L. K., aged 35 years. Ragweed hay fever. Very good results both seasons, 1938 and 1939.

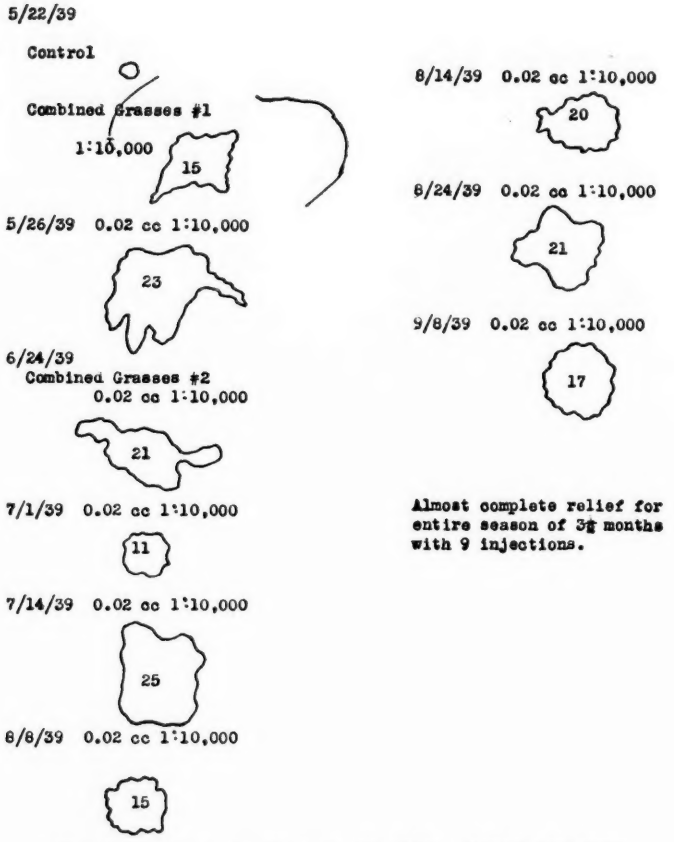


Fig. 7.—Case 6. L. B., aged 20 years. Grass hay fever.

By this method of treatment the size of the skin reaction, the degree of relief, and the intervals between injections indicate the optimum dosage.

In the cases of moderately marked sensitivity a wheal about 15 mm. in diameter is the rule with 0.01 c.c. of the 1:10,000 dilution. The 1:1,000 dilution should not be applied at the first sitting. Treatment should be continued with increasing doses of 0.02, 0.03, 0.04, and 0.05 c.c., and then with 0.01, 0.02, 0.03, 0.04, and 0.05 c.c. of the 1:1,000 dilution according to the plan outlined above. Satisfactory response is usually noted to wheals varying from 20 to 30 mm. in diameter.

In patients with moderate sensitivity a wheal about 15 mm. in diameter is usually obtained with the 1:1,000 dilution. Subsequent injections should be increased by 0.01 c.c. until the proper size wheal is obtained. With the 1:1,000 dilution the amount of the injection may be increased up to 0.05 c.c. or 0.07 c.c.—higher if necessary.

In the patients with less than average sensitivity, the 1:1,000 dilution may be used throughout in doses up to 0.10 c.c. It is rarely necessary to use the 1:100 dilution.

CASE REPORTS

The case reports of ragweed and grass hay fever are presented to illustrate the method of titration of sensitivity and to show the size of the wheals, the amount and dilution of the extracts used, and the intervals between injections.

COMMENT

In the treatment of hay fever of all types and by any method of hyposensitization, satisfactory results are dependent primarily upon (1) the use of pollen extracts to which the patient is sensitive and with which he has contact and (2) adequate or optimum dosage, given at the proper intervals. When these principles of therapy are followed and satisfactory relief is not obtained, it is reasonable to assume that nonpollen sensitivities have not been considered or have not been properly controlled. It is generally known and appreciated that some pure hay fever patients may be sensitive also to common inhalants, such as house dust, occupational dust, orris, feathers, wool, etc., to foods, or to both. Patients who have perennial respiratory symptoms or other manifestations such as gastrointestinal allergy, urticaria, angioneurotic edema, eczema, or allergic headache should be considered as definitely sensitive to nonpollen substances. It is evident, therefore, that in these cases the secondary factors must be adequately controlled.

Fig. 6.—Case 5. L. K., aged 35 years. Ragweed hay fever. [illegible] good results both seasons, 1938 and 1939.

SUMMARY

1. The intracutaneous coseasonal method of treatment of hay fever is adaptable to the tree, grass, or ragweed types.

2. It is especially satisfactory for the tree cases as the pollinating seasons are usually short and irregular.

3. Although the grass season is comparatively long, the counts are never very high and there are usually periods with low counts during which treatment affords protection for longer intervals.

4. The method is most adaptable to small children, thus eliminating a long series of preseasonal or perennial injections.

5. By following the preliminary plan of titration of skin sensitivity unusually large reactions can be avoided. Constitutional reactions are not common.

6. In a few cases better results with each succeeding year of treatment have been noted.

REFERENCES

1. Duke, W. W.: Allergy, Asthma, Hay Fever, Urticaria and Allied Manifestations of Reaction, St. Louis, 1926, The C. V. Mosby Co.
2. Phillips, 9. W.: Relief of Hay Fever by Intradermal Injections of Pollen Extract, J. A. M. A. **86**: 182, 1926.
3. Coca, A. F., Walzer, M., and Thommen, A. A.: Asthma and Hay Fever in Theory and Practice, Springfield, 1931, Charles C. Thomas.
4. Anderson, J. M.: Small Dosage Treatment for Hay Fever, J. ALLERGY **3**: 306, 1932.
5. Anderson, J. M.: A New Method of Preparation of Pollen Extracts, J. ALLERGY **6**: 244, 1935.
6. Phillips, E. W.: Intradermal Pollen Therapy During the Attack, J. ALLERGY **5**: 29, 1933.
7. Matzger, E.: Notes on Coseasonal Pollen Therapy, Internat. Corresp. Club of Allergy **3**: 23, 1940.
8. Dutton, L. O.: Co-Seasonal Treatment, ibid. **3**: 29, 1940.
9. Harris, L. H.: Results of Coseasonal Therapy, ibid. **2**: 99, 1939.
10. Davidson, H. M.: Results of Coseasonal Therapy, ibid. **3**: 16, 1940.
11. Seyler, L. E.: Satisfactory Results With Co-Seasonal Therapy, ibid. **3**: 20, 1940.
12. Brown, O. H.: Hay Fever Therapy, ibid. **3**: 29, 1940.
13. Barnes, M. C.: Impressions on Co-Seasonal Pollen Therapy, ibid. **3**: 37, 1940.
14. Schonwald, P.: Coseasonal Treatment of Pollinosis, ibid. **3**: 47, 1940.
15. Halpin, L. J.: Co-Seasonal Pollen Therapy, ibid. **3**: 49, 1940.
16. Wittich, F. W.: Late Preseasonal and Coseasonal Therapy, ibid. **3**: 43, 1940.
17. Vaughan, W. T.: Practice of Allergy, St. Louis, 1939, The C. V. Mosby Co.

DISCUSSION

DR. I. STANLEY KAHN, San Antonio.—I can confirm the excellent results secured by Dr. Hansel in this method of treatment. The determination of dosage by the securing and maintenance of a wheal of a definite size (finger to thumbnail) is also ordinarily a very satisfactory technique. However, as a result of about fifteen years' experience with this method of treatment used almost daily, at least in my hands, several considerations seem necessary for its success. First, while underdosage means merely inefficacy and wasted time, overdosage, especially with the initial dose which results in any appreciable increase of symptoms, when repeated usually makes subsequent treatments by this method extremely difficult and at times impossible. The determination and application of the proper initial dose almost seem to be the keys to the whole situation.

For any kind of consistent results, two other factors besides size of the wheal must be considered. One is the antigenicity of the particular extract or extracts used, a matter easily learned by experience. For instance, if one is using material with no particular or even a high degree of antigenicity, it is perfectly safe to secure a wheal which is larger than a standard wheal. With material like ragweed of a high degree of antigenicity, these wheals described by Dr. Hansel seem to me about correct.

Also, much more important is the individual clinical sensitivity of a patient, together with the atmospheric antigen dosage received at the time.

Since patients giving negative tests intradermally to ordinary dilution strengths respond to this method and since clinical sensitivity does not invariably parallel cutaneous sensitivity, but may far exceed it, this factor of clinical sensitivity should be given the fullest consideration in determining the potential initial dosage, rather than the routinely and mechanically raising of serial dilutions to a point that produces a reaction of a predetermined necessary size. Such a dose may be and at times is appreciably excessive. No dose should be given beyond this point of apparent clinical sensitivity, even if the wheal reaction be only minimal or even absent. Also, conversely, the actual proper relieving dose may be one that produces a cutaneous reaction of apparently dangerous extent. All this is due to the fact that an intradermal injection represents a constitutional as well as a local dosage and, in my experience, more or less like that of Dr. Gay, one of far greater intensity than a subcutaneous dose of identical size. Consideration must be given that this is coseasonal and not preseasonal therapy.

Guides to the degree of clinical sensitivity existing are the presence or absence of preliminary positive dermal tests, the date of onset of symptoms following the date of seasonal pollen onset, the severity of symptoms when the patient is first seen, and the severity over a number of previous days with relation to the atmospheric pollen contact, as revealed by daily pollen counts over this period.

The factors that govern dosage intervals and increases should again be regulated by the clinical sensitivity, the degree of improvement, and the expectancy of pollen contact. The dosage of a farmer or a man going hunting in a high wind in the middle of a pollen season is entirely different from that of a city downtown worker on a rainy day. Usually at the start, the small doses, in my experience, give a protection of from twenty-four to forty-eight hours' duration. Hyposensitization is rather rapid. Some of these patients will tell you, especially physicians who have some judgment, that a treatment by this method acts almost like an antitoxin, and in one-half to three-quarters of an hour they feel very much better. If the initial wheal-producing dose in these cases is repeated in a half hour in an area 2 or 3 inches distant, the second wheal will come up very, very much more slowly and will be of decidedly smaller size. This calls for persistent dosage increase. In my experience the protection seldom lasts over five or six days, but I have seen it last two or three years.

While this form of therapy often gives brilliant results, its consistent, successful, practical application cannot be based on local reaction alone. To my mind, its use requires an appreciable amount of study and judgment for each patient, far more than is required for determining ordinary routine preseasonal subcutaneous dosages.

It is seldom, in my opinion, a practical routine for a busy clinic or for anyone but an experienced allergist who is willing to give the time and take into consideration pollen counts, wind velocity, and other factors at the time the treatment is given.

There is another point which I think is fairly important. No succeeding dose should be given until all evidence of the preceding reaction has disappeared and cannot be rubbed back by vigorous friction. This means that, when we are handling one of these cases, it may be necessary to obtain telephone reports to the office almost daily for each and every case. I should say it is an extremely difficult method, but a very satisfactory one.

DR. HAROLD A. ABRAMSON, New York.—If one wishes to give either short or giant ragweed or timothy intradermally, an alternate method which might be just as efficacious as the injection technique is the method of electrophoresis. The apparatus used in electrophoresis involves merely a simple galvanic circuit. The extract, which should be salt-free and at pH 7.0 saturates canton flannel and which is placed on the skin of the forearm or thigh of the patient, is introduced by means of the electrical field. Four variables are available to vary the quantity of allergen introduced. The time, the current, the area of the electrode, and the concentration of the solution may be altered.

I cannot at present give an adequate explanation for the mechanism of introduction of allergens. It would seem, however, that, since both poles may introduce the allergen, diffusion processes are just as important, if not more so, than the electrophoretic forces. The positive pole is more efficacious with giant ragweed extract; with dwarf ragweed extract, the negative pole is better. Very marked skin reactions may be obtained if the allergen solution has as much as 0.5 mg. of protein nitrogen per cubic centimeter.

For a given dose the intensity of whealing may be reduced by merely enlarging the area of the electrode. In this way it is likely that skin depots of the allergen are obtained. One patient who has been treated in this way shows a rather interesting type of reaction; delayed whealing occurs three or four hours subsequent to electrophoresis. This wheal may last several days intermittently and lights up if nuts are ingested. Not all people, however, react in that way.

As has been shown elsewhere, the skin is capable of forming depots of histamine. If a molecule as small as histamine can be stored in the pores, I see no reason why molecules much larger cannot be stored in the same way. A simple way of demonstrating the storage of histamine is as follows. Histamine is introduced by electrophoresis using the positive pole. A 1:10,000 dilution of histamine suffices. Applying the positive pole subsequently, even three or four days later, a secondary wheal may appear at the original site of injection. Or by using reversed electrophoresis, histamine may be electrically transported out of the site which originally showed whealing and transferred to another site in the skin using the positive pole. If allergen depots are desired to produce effective immunization, the electrophoretic technique may with further experimentation prove very valuable.

DR. HANSEL.—There are many who have had a great deal of experience with this method of treatment. I am disappointed not to hear

more discussion. This subject was presented primarily with the idea of exciting some discussion, as we should like to know more about the experiences of others with this method.

Dr. Kahn brought out a very important point which should be particularly emphasized and that is the avoidance of overdosage at the outset. If the method of titration of sensitivity is carefully evaluated, there should be less likelihood of overdosage. It is also important to correlate the treatment with the atmospheric counts.

The probabilities are that in the treatment of ragweed patients better results will be obtained in the East than in the Middle West, for in the East the counts are much lower and the response should be better. For example, we have noted that, on the whole, the response in the grass cases is better than in the ragweed cases.

I have had no experience with electrophoresis, but the method Dr. Abramson has described to us is certainly very interesting and worthy of further consideration.

SKIN TESTING METHODS

Multiple Single Sitting vs. Serial Small Group Tests

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HOW many skin tests to do at one sitting is a vexing question. In 1922 Cooke[1] postulated that the summation of the effects of multiple positive skin reactions might result in constitutional or systemic reactions. He stated that he could not cite an example. This postulate was accepted, without confirmatory evidence, by some allergists as a contraindication to multiple skin testing. For example, Coca, Walzer, and Thommen,[2] Tuft,[3] Brown,[4] and Hansel[5] state that more than ten to twenty tests at a time are contraindicated.

Pratt,[6] on the other hand, showed that when either of two antigens alone is capable of producing moderate anaphylactic shock in specifically sensitized guinea pigs, the sum of these antigens in a single dose does not enhance the degree of shock, whereas doubling the shocking dose of either antigen alone results in an increased response. Waldbott,[7] in discussing Pratt's paper, observed that 200 or 300 skin tests are often given at one sitting and only rarely do constitutional reactions occur, despite the fact that many skin tests may be positive. He stated that if there is an accumulative effect of antigens, we should encounter constitutional reactions of skin testing much more frequently.

About ten years ago we began using the intradermal method, cautiously at first with only ten materials, then twenty, then forty. The economic advantage to the patient, the time saved by the physician, and the rarity of systemic reactions encouraged us to continue increasing the number of tests done at one sitting. About five years ago we were giving an average of eighty-one, and for the past four years, 100 to 150 tests at a time. The allergens are lined up serially in trays of loaded syringes before the intradermal tests are started. The average time required for the injections of the 100 to 150 allergens is about eighteen minutes.

The one essential precaution is a preliminary series of scratch tests with about sixty-five of the allergens which commonly give strong skin reactions and those to which the history suggests the patient might be strongly sensitive.

An analysis of the incidence of skin reactions in 2,018 consecutive patients, tested intradermally with from 80 to 150 allergens at one sitting, a total of about 200,000 injections, is presented in Table I.

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Only two patients had systemic reactions. One, early in the series, was tested with the routine allergens, including a 1:1,000 dilution of ragweed. This was an unnecessary repetition and was ill advised, as the preliminary scratch test had been positive for ragweed. The intradermal reaction was huge, spreading over an area about 3 inches in diameter. The systemic manifestations appeared in about thirty minutes. They were characterized by aggravation of hay fever and cough, some tightness in the chest, and moderate lassitude. No urticaria and no significant asthma resulted. The symptoms were not severe enough for her to report them until the next day. In addition to the excessive response to ragweed, there were twelve 1+, five 2+ and 3+, and one average 4+ reactions.

TABLE I

INCIDENCE OF POSITIVE REACTIONS IN 2,018 CONSECUTIVE PATIENTS WITH THE MULTIPLE INTRADERMAL METHOD OF SKIN TESTING*

NUMBER OF POSITIVE TESTS PER PATIENT	0	1-5	6-10	11-20	21-30	Over 30
Incidence of 1+ reactions		518	454	458	264	211
Incidence of 2+ and 3+ reactions		860	171	76	29	6
Incidence of 4+ reactions		450	35	30	3	1
Incidence of all reactions	64	477	414	518	281	335

*In 809 patients, the average number of intradermal tests at one sitting was 81; in 1,209 patients, the average number was about 120.

The other one of the 2,018 patients who showed systemic symptoms had huge local reactions to crabs and shrimp. Generalized urticaria and moderate lassitude followed in about thirty minutes. Five minims of epinephrine relieved her markedly. She was enabled to sleep most of the night by taking two ephedrine and amytal capsules. Some urticaria and lassitude persisted for two days. In addition to the very large reactions to shrimp and crabs, there were seventeen 1+, five 2+ and 3+, and one average 4+ reactions.

Two additional patients had severe constitutional reactions from skin testing. These were not included in Table I because in each case the patient was tested with only one allergen. The case of one patient who nearly died when tested with an extract of an asthma powder has been reported.[8] The other also nearly died when a student tested him, by mistake, with a 1:33 dilution of grass extract instead of a 1:10,000 dilution. In both patients the local reactions were huge and developed rapidly. The constitutional symptoms were severe and appeared quickly.

The common factor in all four of the above instances of constitutional reactions was the huge local reaction, 3 to 8 or 10 inches in diameter. The two more severe reactions appeared sooner than, and

were accompanied by more marked local responses than, the two milder ones. This is typical of constitutional reactions to therapeutic injections of allergens also.

Table I shows that only sixty-four of the 2,018 patients had negative skin tests, whereas 335 had more than thirty positive reactions. One patient, indeed, had over thirty 4+ reactions. One more recent patient, not included in the table, had sixty-nine 1+, forty 2+ and 3+, ten average 4+, and nine negative and doubtful, but no huge reactions. In none of the cases other than those already described was there any distant or systemic disturbance.

The high incidence (96.8 per cent) of positive reactions, compared with 79 per cent and 60 per cent as reported by Feinberg[9] in two small groups, and with 82 per cent as reported by Piness and Miller[10] in a larger group, shows that the extracts were potent. Conversely, the large number of patients, 477, who had only from one to five positive reactions shows that the extracts were not excessively reactive. Bowman[11] showed, in part, that tests placed in the upper portions of vertical lines will cause somewhat stronger responses than those in the lower. The high incidence of positive reactions in Table I suggests that the findings are of greater experimental than clinical value. This will be the subject for a later communication.

To get a somewhat quantitative idea about the cumulative effects of positive skin reactions, fifty skin tests were done on one person. All of a 2 c.c. solution containing 0.3 mg. of histamine was used. This is only 60 per cent of the amount of histamine used in routine gastric analyses. All of the tests were positive. All but a few were graded as 2+, 3+, or 4+. The patient had no systemic reaction except, perhaps, a little transient malaise about forty minutes after the injections. It seems reasonable to infer from this that the amount of histamine-like substance necessary to produce a troublesome systemic reaction is considerably greater than the very small amount necessary to produce fifty average 2+, 3+, and 4+ reactions. It would also seem reasonable to infer that systemic reactions as encountered in routine skin testing are not due to the cumulative effect of any reasonable number of moderately positive reactions.

SUMMARY

No evidence has been found in the literature to support the statements that routine multiple skin testing is more dangerous than a few tests at one sitting.

Evidence is offered that numerous simultaneous average positive skin tests do not cause systemic reactions by their cumulative effects. On the contrary, systemic reactions seem to be associated with one reaction, or a few huge, rapidly developing local reactions, indicative of extreme hypersensitiveness to one or perhaps to a few allergens.

Only two of 2,018 patients, tested with from 80 to 150 allergens at one sitting, had constitutional reactions. Neither of the reactions was alarming. In all, over 200,000 tests were done.

It must be emphasized that one precaution has to be taken before doing multiple skin tests at one sitting. That precaution is a preliminary scratch test with allergens which commonly produce strong skin reactions or to which the history suggests the patient might be strongly sensitive.

REFERENCES

1. Cooke, R. A.: Studies in Specific Hypersensitiveness. III. On Constitutional Reactions: The Dangers of the Diagnostic Cutaneous Test and Therapeutic Injection of Allergens, J. Immunol. **7**: 119, 1922.
2. Coca, Arthur F., Walzer, Matthew, and Thommen, August A.: Asthma and Hay Fever in Theory and Practice, Springfield, 1931, Charles C. Thomas, p. 316.
3. Tuft, Louis: Clinical Allergy, Philadelphia, 1937, W. B. Saunders Co., pp. 89, 92.
4. Brown, Aaron: Personal communication.
5. Hansel, French K.: Allergy of the Nose and Paranasal Sinuses, St. Louis, 1936, The C. V. Mosby Co., p. 298.
6. Pratt, Henry N.: Anaphylaxis in Multiple Sensitive Guinea Pigs, J. ALLERGY **9**: 14, 1937.
7. Waldbott, George: Discussion of Pratt.[6]
8. Swineford, Oscar, Jr.: Hypersensitiveness to Asthma Powders, J. ALLERGY **8**: 607, 1937.
9. Feinberg, Samuel M.: Allergy in General Practice, Philadelphia, 1934, Lea & Febiger, pp. 116-119.
10. Piness, George, and Miller, Hyman: Skin Tests in 4,589 Cases of Allergic Disease, With a Criticism Concerning Elimination Diets, J. ALLERGY **4**: 18, 1932.
11. Bowman, Katherine L.: Pertinent Factors Influencing Comparative Skin Tests on the Arm, J. ALLERGY **7**: 39, 1935.

ORAL IMMUNIZATION FOR POISON IVY DERMATITIS

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MANY methods for immunization against the poison ivy dermatitis have been advocated, but none of them have been entirely satisfactory. It has been recommended that the plant oleoresin be used as a topical applicant, by parenteral administration or as oral medication.

In 1932 Maisel[1] published a brief report on the value of frequent bathing in dilute *Rhus oleoresin* extract in increasing concentration. The single patient so treated was considered to have been successfully desensitized.

Attempts to produce desensitization by the subcutaneous administration of the plant oil have been numerous. A review of the literature has shown this method to be generally unsatisfactory.[2]

As early as 1829, Dakin[3] noted the effects of *Rhus toxicodendron* leaves when taken by mouth. He recorded that many laborers chewed the leaves of the plant and believed that they were thereby protected from acquiring the dermatitis. This procedure was recommended by many physicians at the time.

The American Indian has been said to eat fresh green leaves from the poison ivy plant for purposes of immunization[4] although this was apparently not a universal custom.[5]

Alderson and Pruett[6] stated that it was a common practice in California to eat some of the poison ivy leaves for prophylaxis. In their camp many men made and drank a soup of the leaves with good prophylactic results.

In 1932 Cooper[7] advised giving patients a small fresh leaf of the plant in early spring to chew and swallow. Immunity was said to follow in three days, and subsequent re-immunization for several years resulted in permanent cure. This immunizing procedure had been used several hundred times by the author without a failure.

Schamberg[8] in 1919 advocated the giving of a tincture of *Rhus toxicodendron* orally in ascending dosage with a subsequent daily maintenance dose. Immunity was said to result for one month. Apparently satisfactory results were reported by Templeton[9] following the technique of Schamberg.

Using a triturate of a tincture of *Rhus toxicodendron* by mouth, Spain and Cooke[4] obtained good results in a small series when small daily doses were given for a long time.

The present study was carried out among the vacationists at a large summer camp for boys maintained at Treasure Island, N. J., by the Philadelphia Boy Scout Council. The many natural advantages provided at this camp for such a study as this have been described previously.[2]

Several months before going to camp, a group of boy scouts and their adult leaders, selected at random, were invited to partake of a poison ivy extract as a preventive for the specific dermatitis. The material used was an ether-soluble extract of the poison ivy leaves prepared as a tablet with an enteric coating.* Each tablet was made to contain 0.01 mg. of the ether-soluble plant extract. The enteric coating was designed to prevent spread of the poisonous material around the mouth with the formation of the local dermatitis. It was also intended to prevent gastrointestinal upsets. The dosage recommended by the manufacturer was followed, namely one tablet daily for seven doses, then one twice weekly. This therapy was continued to and throughout the stay in camp. The total number of tablets taken varied in individual cases from fifteen to thirty. Individual case records were carefully kept. During the interval before going to camp, many of the boys (while taking the tablets) acquired the dermatitis from exposure to the plant. One boy who denied such exposure developed on his abdomen a dermatitis which was indistinguishable from the poison ivy dermatitis suffered on previous occasions. No one complained of any untoward effects, such as pruritus, nausea, or proctitis, due to the medication.

TABLE I

GROUP	NO. PERSONS	NO. CASES OF P.I.	PER CENT P.I.
Experimental	100	32	32.0
Control	1,295	471	36.4

TABLE II

GROUP	NO. PERSONS KNOWN SUSCEPTIBLES	NO. CASES OF P.I.	PER CENT P.I. IN KNOWN SUSCEPTIBLES
Experimental	53	28	52.8
Control	578	328	56.8

There were 100 persons in the experimental group. This does not include many other cases where therapy was not carefully followed as prescribed. Of these 100 persons, thirty-two (32.0 per cent) developed poison ivy dermatitis during their camp sojourn. A control group of 1,295 persons studied simultaneously at the same camp showed 471 persons (36.4 per cent) to have acquired the dermatitis in the same interval.

Of the 100 persons in the experimental group, fifty-three were known to be susceptible to the dermatitis by their past history. There

*Obtained through the courtesy of Sharp and Dohme, Mulford Biological Laboratories.

were twenty-eight cases (52.8 per cent) of poison ivy dermatitis among this known susceptible group, all having acquired it during the camp period studied. In the comparable control group of 578 persons, 328 (56.8 per cent) were afflicted during the same interval.

TABLE III

GROUP	NO. PERSONS, SUSCEPTIBILITY UNKNOWN	NO. CASES OF P.I.	PER CENT P.I. IN PERSONS, SUSCEPTIBILITY UNKNOWN
Experimental	47	4	8.5
Control	717	143	19.9

It would seem that the material used in the manner described was not particularly effective in preventing the dermatitis in susceptible persons following exposure. It may be that more frequent administration would be indicated. If so, it is important to bear in mind that violent gastrointestinal disturbances and proctitis may follow overdosage.[3, 10]

SUMMARY

An attempt was made to immunize 100 persons against the poison ivy plant by the oral administration of the plant extract in enteric coated tablets.

The results were considered unsatisfactory inasmuch as there was practically the same percentage of poison ivy dermatitis cases among the treated persons as among a large group of controls.

REFERENCES

1. Maisel, F.: Poison Ivy—New Method of Immunization, J. ALLERGY **4**: 35, 1932.
2. Zisserman, L., and Birch, L.: Prophylaxis and Treatment of Poison Ivy Dermatitis With an Extract of Rhus Toxicodendron, J. ALLERGY **10**: 596, 1939.
3. Dakin, R.: Poison Ivy, Am. J. M. Sc. **4**: 98, 1829.
4. Spain, W. C., and Cooke, R. A.: Studies in Specific Hypersensitiveness, J. Immunol. **13**: 93, 1927.
5. Gilmore, M. R.: Uses of Plants by Indians of Missouri River Region. Thirty-third Annual Report Bureau American Ethnology 1911-12, Washington, D. C., 1919, Gov. Printing Office, p. 100.
6. Alderson, H. E., and Pruett, H. J.: Poison Oak Dermatitis, California State J. Med. **19**: 188, 1921.
7. Cooper, H.: Immunization for Rhus Toxicodendron Poisoning, M. Rec. **136**: 297, 1932.
8. Schamberg, J. F.: Desensitization of Persons Against Ivy Poison, J. A. M. A. **73**: 1213, 1919.
9. Templeton, H.: Untoward Reactions Following Toxin Treatment for Rhus, Arch. Dermat. & Syph. **20**: 83, 1929.
10. McNair, J. B.: Ivy Poison, Chicago, 1923, Chicago University Press.

A COMPARATIVE STUDY OF THE INCIDENCE OF ACUTE INFECTIOUS DISEASES IN ALLERGIC AND NONALLERGIC PERSONS

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THE questions whether adults suffering from allergic diseases have a super-resistance to pathogenic organisms and whether the allergic child is exceptionally free from the usual diseases of childhood have been controversial.[1] In the literature one finds a wide diversity of opinions among allergists. Balyeat[2] stated that for several years he had noted from the histories he had taken of patients with asthma and hay fever that 93 per cent reported that other than their hay fever or asthma and the diseases of childhood they had had no illness. They usually added that these diseases, when present, were very mild. Balyeat found it uncommon for frail looking persons to seek treatment for allergy excepting those who had had asthma over a long period of time.

Bray[3] concluded that allergic persons are particularly free from other ill-health and are above normal mentally. Rackemann and Colmes[4] were of the opinion that allergic individuals are more susceptible to tuberculosis, colds, and acute infectious diseases than are normal people. Walzer[5] stated that it is noteworthy that, with the exception of infections of the upper respiratory tract, there are few other infections with which asthma is commonly associated. Duke,[6] however, believed that patients with perennial hay fever and asthma are predisposed to acute infections of the respiratory tract, such as coryza and bronchopneumonia. He considered it possible that this is a result of mechanical obstruction in the sinuses and bronchial tree, resulting from the swollen membranes. He believed, however, in addition to this, that chronic reactive patients are immunologically different from normal persons and are actually predisposed to acute infectious diseases. He stated that patients with chronic allergy have a relative immunity to chronic infections such as syphilis and tuberculosis and in them these diseases run a remarkably favorable course. Feinberg[7] stated that it has been noted by many that fewer acute infectious diseases occur in the asthmatic group than in control groups. Urbach[8] claimed that pneumonia appears uncommonly frequently in the histories of asthmatic patients. Coke[9] noted that infectious diseases probably occur with no greater frequency than would appear in the history

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of any child, whether asthmatic or not. Vaughan[10] was of the opinion that, usually, allergic persons are otherwise in remarkably good physical condition. "They may suffer from any of the diseases which attack nonallergic persons, but, as a rule, were it not for their allergy, they would have no complaint. This may be more apparent than real, and further studies are needed. Formerly it was taught that asthma protected the individual to a great extent against tuberculosis, but in recent years many asthmatic patients have been found to have the disease."

TABLE I

FREQUENCY DISTRIBUTION OF CHILDHOOD DISEASES IN ALLERGIC AND NONALLERGIC PERSONS

NUMBER OF AILMENTS	0	1	2	3	4	5
Allergic	8	19	38	38	95	22
Nonallergic	21	14	45	56	91	23

TABLE II

FREQUENCY DISTRIBUTION OF OTHER ACUTE INFECTIOUS DISEASES IN ALLERGIC AND NONALLERGIC PERSONS

NUMBER OF AILMENTS	0	1	2	3	4	5	6	7	8	9	10	11	12
Allergic	4	5	8	28	41	54	60	25	15	5	4	1	0
Nonallergic	5	7	16	22	45	47	57	24	14	7	3	2	1

To clarify this point a survey has been made of 500 adults. Of these, two hundred and fifty were definitely allergic: sixty-seven complaining primarily of asthma, two of abdominal allergy, three of angioneurotic edema, forty-four of allergic dermatitis, five of eczema, forty of hay fever, seven of food allergy, twenty of migraine, thirty-seven of vasomotor rhinitis, one of vernal catarrh, and twenty-four of urticaria. One hundred and ninety-one of the allergic patients complained of the presenting condition only, while forty-five had two allergic complaints, twelve had three, one had four, and one had five. The other 250 had always been free from allergic conditions. All were carefully questioned concerning the incidence of childhood diseases (measles, mumps, chicken pox, whooping cough, scarlet fever, diphtheria), and other acute infectious diseases not confined to childhood (specifically, influenza, tonsillitis, chorea, inflammatory rheumatism, pleurisy, pneumonia, malaria, typhoid, dysentery [amebic or bacillary], syphilis, gonorrhea, and also the details of any other diseases the patient might mention). The frequency distribution of childhood diseases in these allergic and nonallergic persons is given in Table I. Table II shows the frequency distribution of all infectious diseases suffered by allergic and nonallergic persons, and the statistical analysis indicates that these differences are without significance.

The mean number of ailments suffered by the allergic persons is $3.156 \pm 2(0.076)$, and the standard deviation is 1.205. The mean number of

ailments suffered by nonallergic persons is 3.004 ± 2(0.086), with a standard deviation of 1.375.*

The mean of the allergic persons is 5.32 ± 2(0.1120), and the standard deviation is 1.870. The mean of the nonallergic persons is 5.112 ± 2(0.1269), and the standard deviation is 2.006.*

TABLE III

DETAILED INCIDENCE OF ACUTE INFECTIOUS DISEASES IN ASTHMATIC, NONASTHMATIC ALLERGIC, AND NONALLERGIC PERSONS

DISEASE	ASTHMATIC (67)		OTHER ALLERGIC (183)		NONALLERGIC (250)	
	NO. OF CASES	PERCENTAGE	NO. OF CASES	PERCENTAGE	NO. OF CASES	PERCENTAGE
Measles	59	88.0	175	95.0	230	92.0
Mumps	40	57.9	111	60.6	168	67.9
Chicken pox	36	53.8	121	66.1	141	57.6
Whooping cough	49	73.1	131	71.5	174	69.6
Scarlet fever	12	18.3	20	10.9	20	8.0
Diphtheria	6	9.1	10	5.4	24	9.6
Influenza	47	70.1	124	67.9	160	64.2
Tonsillitis	25	37.3	90	48.6	115	46.5
Pneumonia	18	27.4	36	19.9	36	14.4
Smallpox	4	3.0	2	1.0	8	3.2
Typhoid	11	8.3	22	12.0	41	16.4
Inf. rheumatism	2	1.5	8	4.3	14	5.6
Malaria	12	18.3	34	18.5	68	25.2
Bacillary dysentery	5	3.7	11	5.9	19	7.6
Gonorrhea	2	1.5	10	5.4	16	6.4
Meningitis			1	0.5		
Syphilis			4	2.0	2	0.8
Erysipelas			2	1.0		
Pyelitis			1	0.5	2	0.8
Pleurisy	14	20.9	8	4.3	34	13.6
Chorea	3	2.3	1	0.5	1	0.4
Encephalitis	1	0.8				
Tuberculosis	1	0.8			2	0.8
Herpes zoster	1	0.8			1	0.4
Amebiasis	1	0.8				
Acute nephritis					2	0.8
Colitis					1	0.4
Ethmoiditis					1	0.4

Indeed, the widespread incidence of allergy reported by numerous observers would tend to bear this out on a priori grounds. The similarity in incidence of infectious processes in the two groups corroborates the statement of Vaughan[14] that there is no fundamental dif-

*If the allergic person has a mean of 3.156 ± 2(0.076) childhood ailments and 5.32 ± 2(0.1120) nonchildhood infectious diseases as contrasted with means of 3.004 ± 2(0.086) and 5.112 ± 2(0.1269) in nonallergic persons, the question arises as to whether these differences are significant. The standard error of the difference between two means may be computed by the formula:[11]

$$\sqrt{\frac{\sigma_1^2}{N_1} + \frac{\sigma_2^2}{N_2}} = \text{Standard error of difference}$$

When σ_1 = standard deviation of first group
σ_2 = standard deviation of second group
N_1 = number in first group
N_2 = number in second group

To be significant the difference must be at least twice this value or it might have arisen by chance. The standard deviation of the mean for childhood diseases in allergic persons is 1.205 and in nonallergic persons is 1.375. Substituting, we find that the standard error of the difference between the means is 0.1150. In a similar manner, the standard error of the difference between the means of all infectious diseases may be calculated. The standard deviation of the allergic persons is 1.870, and of the nonallergic, 2.006. Substituting in the formula we get a result of 0.1735.

ference between the allergic and the so-called nonallergic individual. Even earlier, Doerr[12] expressed his belief that the capacity to become sensitized is present in all human beings, differing only in degree. Finally, attention is called to the work of Rackemann and co-workers,[13] who found no fundamental difference in the development of typhoid and other agglutinins in allergic and nonallergic persons.

SUMMARY

In the series studied, no significant difference was found in the incidence of acute infectious diseases in allergic and nonallergic persons.

REFERENCES

1. Balyeat, R. M.: The General Health and Mental Activity of Allergic Children, Am. J. Dis. Child. **37**: 1193, 1929.
2. Balyeat, R. M.: Hay Fever and Asthma, ed. 2, Philadelphia, 1928, F. A. Davis Co.
3. Bray, G. W.: Recent Advances in Allergy, ed. 2, Philadelphia, 1934, P. Blakiston's Son & Co., p. 208.
4. Rackemann, F. M., and Colmes, A.: Studies in Asthma. V. The Clinical Characteristics of Hypersensitiveness, J. ALLERGY **1**: 2, 1939.
5. Coca, A. F., Walzer, M., and Thommen, A. A.: Asthma and Hay Fever in Theory and Practice, Springfield, Ill., 1931, Chas. C. Thomas.
6. Duke, Wm. W.: Allergy, Asthma, Hay Fever, Urticaria, and Allied Manifestations of Reaction, St. Louis, 1925, The C. V. Mosby Company, p. 68.
7. Feinberg, Samuel M.: Allergy in General Practice, Philadelphia, 1934, Lea & Febiger, p. 46.
8. Urbach, Erich: Klinik und Therapie der Allergischen Krankheiten, Vienna, 1935, Verlag Wilhelm Maudrich, p. 357.
9. Coke, Frank: Asthma, ed. 2, Baltimore, 1939, Williams & Wilkins Co., p. 44.
10. Vaughan, Warren T.: Allergy and Applied Immunology, ed. 2, St. Louis, 1934, The C. V. Mosby Co., p. 293.
11. Hill, A. B.: Principles of Medical Statistics, Lancet **1**: 1937.
12. Doerr, R.: Die Anaphylaxieforschung, Ergebn. d. Hyg., Bakt., Immunitätsforsch. u. exper. Therap. **5**: 71, 1922.
13. Rackemann, F. M., Simon, F. A., Simon, M. G., and Scully, M. A.: Further Observations on the Nature of Allergy, J. ALLERGY **4**: 498, 1933.
14. Vaughan, Warren T.: Practice of Allergy, St. Louis, 1939, The C. V. Mosby Co., p. 110.

FLOUR ALLERGY AND EPITHELIAL HYPERSENSITIVENESS TO AMMONIUM PERSULFATE IN BAKERS AND MILLERS

H. A. E. v. Dishoeck, M.D., Amsterdam, and D. J. Roux, M.D.

HYPERSENSITIVENESS to flour and to persulfate in bakers and millers is a subject that holds the interest of rhinologists,[1] dermatologists, allergists, and hygienists. The large number of bakers and millers suffering from nasopharyngitis, vasomotor rhinitis, asthma, and eczema, as well as the hygienic preparation of bread and the resulting loss of employment by the unfit worker, make this problem of considerable medical and social importance.

Zitzke,[2] Colmes et al.,[3] Hlaváček,[4] Baagoë,[5] v. Vonno and Struycken,[6] and Prakken and Postma,[7] have contributed valuable work on illnesses due to flour. The results of these researches and the conclusions one must draw from them contradict one another. Flour hypersensitiveness has been shown by many investigators, not as one would expect, however, in approximately all patients with allergic symptoms who work with flour, but only in ±50 per cent. Some doubt arose as to the importance of the flour as an allergen. This doubt was strengthened by the fact that dermatologists, such as Zitzke and Prakken, could show a hypersensitiveness to the ammonium persulfate with which flour is treated in nearly all their patients suffering from active bakers' eczema. In this way, the view arose, that bakers' eczema is due to a persulfate hypersensitiveness, to which a hypersensitiveness to flour is added in some cases (Zitzke, Prakken and Postma). Another view was that the flour improvers played no part in the origin of bakers' eczema and that the sensitization to flour was the chief factor (v. Vonno and Struycken). The sensitizing action of lactic acid, which develops in fermenting dough, was shown by Colmes and Guild and Rackemann in fourteen of thirty-two bakers with eczema.

It appeared to us in the course of this investigation that the method of research and the restricted selection of patients was responsible for these fundamental differences. Thus, the rhinologist sees cases of vasomotor rhinitis. The dermatologist, on the other hand, sees chiefly patients with active bakers' eczema and usually tests them by the patch test method, in which importance is attached to chemicals which are added to the flour.

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With extracts that contain several allergens and no irritating substances, the reactions by the method of intracutaneous injection are very sensitive and suitable for quantitative examination. But when one uses extracts from ordinary flour, many nonspecific irritating substances, including histamine, are brought into the skin. These substances in some cases produce pseudoreactions where no hypersensitiveness to flour exists, while in others, the quantity of specific allergens is sometimes too small to show an existing hypersensitiveness to flour. As a result, as shown by different investigations, numerous pseudoreactions appeared in control cases, and negative reactions repeatedly occurred in patients with flour hypersensitiveness.

In our investigations we used the scratch test with very pure proteins obtained from various kinds of grain. This method causes the patient the minimum amount of discomfort, is not dangerous, and gives a reaction which can be clearly seen; the result can be read within fifteen minutes, and pseudoreactions in control cases do not occur. A general allergic examination of the patient was made also.

TABLE I

HYPERSENSITIVENESS TO FLOUR WITH THE SCRATCH METHOD IN CONTROL CASES

NO. OF PATIENTS	TYPE	POSITIVE FLOUR REACTIONS
150	Test persons (not bakers)	1 (gardener)
35	Allergic patients (not bakers; no hay fever)	0
20	Eczema patients (not bakers' eczema)	0

As bread in Holland is prepared entirely from wheat (only cake bakers use other kinds of flour mixtures), one would expect that all sensitized flour workers would show a positive wheat reaction. That, however, was not the case. The number of patients hypersensitive to wheat was considerably greater than those hypersensitive to other kinds of flour, but the extraordinary fact appeared that some patients, suffering from contact with flour, showed strong reactions to oat only. They seldom, if ever, came in contact with this flour, but were in daily contact with wheat. Of seventy patients hypersensitive to flour, fifty-five gave wheat reactions. Had we (like some investigators) restricted our investigations to wheat flour only, we would have obtained negative results in 21 per cent of our cases. Since we also tested with other kinds of flour, especially oat and rye, we succeeded in obtaining positive reactions in these patients.

To determine hypersensitiveness to persulfate, the patch test method with 1 per cent and 5 per cent solutions, according to Zitzke, is the most suitable. This concentration is much stronger than that in the flour, viz., 0.01 per cent. The results can be seen after one or even several days. By intracutaneous injections and by the scratch test

method, we so often saw irritating phenomena, as did v. Vonno and Struycken, even in normal people, that we did not apply this method further.

TABLE II

HYPERSENSITIVENESS TO AMMONIUM PERSULFATE WITH THE PATCH TEST METHOD

NO. OF PATIENTS	TYPE	POSITIVE PERSULFATE REACTIONS
50	Test persons	0
50	Allergic patients	0
10	Eczema sufferers (not bakers)	0

In addition we have examined 178 healthy flour workers, i.e., 81 bakers and 97 millers. All these reacted negatively to the different varieties of flour and to ammonium persulfate. Therefore, it seems evident that the sensitization to flour and persulfate is in no way an attribute of the healthy flour worker. In normal persons and in allergic patients, who are not bakers, flour hypersensitiveness seldom occurs.

On the other hand, among the flour workers, those who were suffering or who had suffered from vasomotor rhinitis, asthma, or eczema, we found, as shown in Table III, positive flour reactions in a very high percentage of cases, but persulfate hypersensitiveness in only one-third of the cases suffering from active eczema.

TABLE III

HYPERSENSITIVENESS IN FLOUR WORKERS WITH SYMPTOMS OF ALLERGY

OCCUPATION	ILLNESSES	NUMBER	FLOUR REACTIONS (SCRATCH TEST)		PERSULFATE REACTIONS (PATCH TEST)	
			+	-	+	-
Bakers	Respiratory tract	47	43	4	0	47
	Eczema present	33	33	0	11	22
	Eczema in past	21	9	12	0	21
Millers	Respiratory tract	9	9	0	0	9
	Eczema present	8	6	2	0	8

The problems presented by the four bakers with respiratory allergy and negative flour reactions (Table III) were solved in three cases. Another allergen was found to be the probable cause of the vasomotor rhinitis and asthma (e.g., cats, eggs, milk). When these exceptional cases are left out of consideration, one can conclude that symptoms caused by flour—vasomotor rhinitis, asthma, eczema, etc.—usually go hand in hand with positive skin reactions to flour proteins.

The thirty-three bakers, who were suffering from eczema during our investigations, were without exception strongly hypersensitive to flour and one-third of them to persulfate. If we had examined a larger number of bakers, there might have been, as among the millers,

an eczema patient with negative flour reactions, that is to say, a baker with some skin affection other than bakers' eczema. The reverse could also occur. For example, a psoriasis sufferer, who is a baker, may be sensitized by the flour, and we may thus find the combination of a hypersensitiveness to flour and a skin disease which is not bakers' eczema. That these cases occur is shown to be possible by our examination of twenty-one bakers who had been suffering from eczema, some of them a few, others many, years ago. Twelve workers among them were not sensitive to flour when examined. Among these twelve, five patients had suffered, in localization as well as progress, from a very atypical eczema which probably was not bakers' eczema. In the other cases, it is possible that, in the course of years (3 to 25), the hypersensitiveness to flour was lost. Among the eight millers who suffered from eczema during our investigation, we actually found one patient with eczema on the elbow and one with eczema on the buttocks, neither of whom were hypersensitive to flour. The connection of the eczema of these patients with their trade was very dubious. The other six patients showed positive reactions to flour.

The hypersensitiveness to persulfate was entirely an attribute of the skin of bakers suffering from eczema and was always accompanied by a hypersensitiveness to flour. An isolated hypersensitiveness to persulfate, although one should admit the possibility, we have never found. On the other hand, we often found a hypersensitiveness to flour without a persulfate hypersensitiveness. Whereas Zitzke and the other investigators state that in some cases the hypersensitiveness to flour develops secondary to that to persulfate, our investigation proves very much to the contrary, namely, that among those sensitized to flour, a hypersensitiveness to persulfate sometimes develops temporarily. Thus, one finds among flour workers an eczema with flour hypersensitiveness and without persulfate hypersensitiveness, which one could call an allergic flour eczema. One also comes in contact with this eczema in countries where no flour improvers are used (Switzerland). Apart from this, one finds patients with eczema who show a flour, as well as a persulfate. hypersensitiveness. The clinical aspect of this eczema is somewhat different from that of the allergic flour eczema. So it is possible in selecting these cases and testing them with persulfate for a very high percentage of positive skin reactions to be found.

REFERENCES

1. v. Dishoeck, H. A. E., and Roux, D. J.: Meelziekten, Bussum, 1938.
2. Bering, F., and Zitzke, E.: Die beruflichen Hautkrankheiten, Leipzig, 1935, Leopold Voss.
3. Colmes, A., Guild, B. T., and Rackemann, F. M.: J. ALLERGY **6**: 539, 1935.
4. Hlaváček, V.: Acta oto-laryng. **26**: 358, 1938.
5. Baagoë, K. H.: Acta med. Scandinav. **27**: 80, 1933.
6. V. Vonno, N. C., and Struycken, J.: Acta dermat.-venereol. **15**: 227, 1934.
7. Prakken, J. R., and Postma, C.: Nederl. tijdschr. v. geneesk. **82**: 367, 1938.

ASTHMA FROM INSECT EMANATIONS

Report of a Case Due to Moths

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IN CONTRAST to the emanations of certain animal species, insect emanations seem to be responsible for allergic manifestations comparatively rarely. Of the twenty-three important orders of insects[1] only three would appear at present to be of major importance in this respect: the *Lepidoptera* (moths and butterflies), *Trichoptera* (caddis flies), and *Ephemerida* (May flies). The first two are characterized by wings covered on both surfaces by scales or hairs of varying shape, which are easily rubbed off, either in flight or on the most gentle contact, forming a fine "dust" which is readily wind-borne[1] (Fig. 1).

Fig. 1.—Portion of the wing of the common or webbing clothes moth, showing the scales and hairs which cover its surface (×160).

Such desquamations are demonstrated without difficulty both grossly and microscopically[2, 3] (Fig. 2). It is stated[2] that "more scales and hair are cast into the air by a single moth than the amount of pollen which is blown from the stamen of a timothy grass over a similar period of time." In such widely separated localities as Buffalo,[2] Florida[4] and Oklahoma City,[5] moth scales and hairs have been identified on slides exposed during the summer—on some days, in numbers exceeding that

From the Jewish Hospital, Philadelphia; service of Dr. Erich Urbach.

of all pollens combined. Slides exposed in seven cities in four mid-western states and Canada, as well as up to 200 feet above street level, have shown the presence of caddis fly hairs.[6]

Fig. 2.—Scales and hairs brushed from the wing of a common clothes moth (×160).

With May flies, the mechanism differs in that no insect "dust" is scattered. However, dried fragments of the thin, delicate pellicle shed by the insect after the subimago stage of its life cycle are readily windborne and exceedingly abundant in certain vicinities.[7]

Other insects, such as house flies and bees, lack these characteristics and hence are much less frequently mentioned as causing symptoms of hypersensitiveness by inhalation of insect fragments.

This paper is devoted exclusively to insect emanations, and therefore excludes from consideration other forms of insect allergy, such as bites and stings of flies, fleas, mosquitos, and bees. It should also be pointed out that we are using the term insect in its strict entomologic sense and are so excluding mites, which in their dead and disintegrated state, are often responsible for asthma.

We believe the case below to be of interest because it is the first reported of asthma due to the common or webbing clothes moth (*Tineola biselliella* Hummel). It is also the first reported case of moth allergy in which there is no element of occupational exposure.[2, 8-10] It differs from many cases of insect allergy in that there is no seasonal incidence. Finally, excellent results have been obtained by hyposensitization treatment.

LITERATURE

In 1918 Caffrey,[8] an entomologist working with the New Mexico range moth (*Hemileuca oliviae* Cockerell), observed in himself and several coworkers, that continued contact with the spines of the larvae and the hairs of the adults produced first hay fever-like symptoms and then violent paroxysms of coughing and wheezing. These observations were later confirmed by Randolph,[9] who reported similar symptoms in an

entomologist due to inhalation of the dust from the floor of the cage in which these insects were kept. Positive intradermal tests were obtained with extracts of the dust and of the insect eggs, and successful passive transfer tests were performed, indicating the allergenic nature of the coverings and spines of the larvae and the material left behind in the egg shells after the emergence of the larvae. It is interesting to note that the spines of this caterpillar have long been known to produce urticarial wheals.

Parlato,[2] in 1932, pointed out the allergenic nature of the emanations of the insect order *Lepidoptera* (moths and butterflies), which is biologically closely related to caddis flies,[1] and like them has epithelial setae which are readily desquamated. A case of seasonal hay fever presumably due to exposure to moths and butterflies while the patient was gardening was reported. The diagnosis was based on the large number of hairs and scales seen on glass slides exposed in the patient's home, on the negativity of routine tests, and on the absence of symptoms when the patient was elsewhere. Skin tests for insect allergens were not performed in this patient. While preparing these insects for extraction, Dr. Parlato's office technician was troubled by tickling of the nose and paroxysms of sneezing and was found to have positive skin, conjunctival, and passive transfer tests to them.

Extracts of the wings of "several species" of moths and butterflies were tested by Parlato in six cases of caddis fly hypersensitiveness, with positive skin and eye reactions in four and moderate skin reactions in two. On the basis of this observation and of an extensive series of "exhaustion" studies of passively sensitized skin sites,[11] Parlato concluded that the caddis fly antibody is similar to that of the moth and butterfly, while that of the last two are identical.

Wittich,[10] in reviewing occupational exposure to allergens in grain and seed mills, found that the Indian meal moth (*Plodia interpunctella* Hbn.) of the order *Lepidoptera* produces respiratory allergy due to the heavy infestation of shelled seed corn with the epithelium of their wings.

In summary, only two particular species of moths (the New Mexico range moth and the Indian meal moth) have been specifically indicted as causing sensitization, both as a result of occupational exposure.

Of the other insects, the caddis fly and the May fly have received considerable attention. Parlato[3] reported, in 1929, the first case of coryza and asthma due to the hairs and scales of caddis flies, also known as sand flies, comprising the order *Trichoptera*. Skin and conjunctival tests, passive transfer, and clinical exposure were all positive. Subcutaneous injections of an extract resulted in successful hyposensitization. He[12] has since recorded a total of forty-three cases of asthma and hay fever, including nine also troubled by hives and eczema, due to this cause. Also in 1929, Figley[7] reported four cases of seasonal asthma,

three of them associated with hay fever, due to May flies *(Ephemerida)*, variously known as shad, lake, or river flies. All the patients had strongly positive skin tests, and one was satisfactorily treated pre- and coseasonally. One patient also demonstrated huge hives after direct contact with the flies. MacDermot[13] described a similar case of asthma. Parlato[14] found nineteen positive skin reactions to the May fly in a group of 589 cases of seasonal hay fever and asthma. Of these, seven yielded positive ophthalmic tests, and six, positive passive transfer. Three cases were successfully treated by pre- and coseasonal methods. Figley[15] has recently reported forty cases of seasonal hay fever and asthma owing all or part of their symptoms to the May fly. Results of hyposensitization were quite satisfactory. Unger[16] reported that almost every worker in power plants along the Mississippi river eventually becomes sensitized to "river flies," developing rhinitis or asthma and that symptoms are relieved by injections of an extract of the insects.

A variety of other insects have been held responsible for isolated cases of allergy. Sternberg[17] reported a case of asthma due to contact with bedbugs, with positive reactions and passive transfer. Here, however, the element of insect bites is not ruled out. Two cases of asthma due to the emanations of bees were proved by Ellis and Ahrens.[18] Weil[19] mentions a case of hay fever due to the dead bodies of the *tanytarsus*, a midge of the family *Chironomidae*, in a hydroelectric dam worker in Alabama. Asthma resulting from sensitization to the house fly (*Musca domestica*) has been reported by Jamieson[20] and to the mushroom fly (*Aphiochaeta agarici*) by Kern.[21] Wittich[10] found positive skin reactions to extracts of book lice (*Troctes divinatorius* Mull) and two cases of allergic rhinitis and asthma[22] due to the Mexican bean weevil (*Zabrotes subfasciatus* Boh.) in grain and seed mill workers.

CASE REPORT

A. B. I., white, aged 11 years, has had recurrent attacks of typical asthma since the age of 2 years, following pertussis. Symptoms were present only in his family home in New York, and never in any other house, or on trains, or in several hospitals, or outside New York. Attacks occurred approximately once a month and were somewhat more frequent at night. Wind, cold, and dust were not noted to have any effect, but emotional excitement was observed to have some influence on the onset of attacks. Nasal obstruction was present but only when accompanied by the asthma.

A diagnosis of house dust asthma had been made some years previously, but treatment with autogenous dust extract had little effect. At the age of 6 years, he was sent to Arizona for seven months, and during this time he was free from symptoms, but the asthma recurred shortly after he returned home. Increasingly severe attacks again required his residence in Arizona for two full years. However, eight days after he again reached home, his condition had become so distressing that one of us (E. U.) was consulted.

The patient's mother stated that, since its purchase fourteen years before, their home had been heavily infested with moths, which resisted all attempts to control

them, including the efforts of professional exterminators. In fact, only eight days before the patient was first seen, a mattress had been opened and found to be "full" of moths.

Past History.—Aside from pertussis, the patient had suffered from chicken pox, measles, and mumps in his earlier years. He was not at all subject to colds or bronchitis. Tonsillectomy and adenoidectomy had been performed at the age of 5 years, but apparently had not affected the course of his disease.

Family History.—The patient's father had specific asthma due to cats, and also spring and fall hay fever. His sister also had hay fever. No other allergies were known to exist in members of the family.

Physical examination revealed a normally developed, intelligent boy with the classical signs of asthma on his first visit. These disappeared almost completely after twenty-four hours of hospitalization, and entirely so after forty-eight hours.

Tests.—Scratch test with powdered webbing clothes moth gave a strongly positive urticarial reaction with a marked halo, but was negative on a control (patient's father). With coddling moth, scratch test (1:100 dilution) was negative; intracutaneous test (1:1,000 dilution) produced a moderate wheal with a marked halo, but was negative on a control (mother). Other skin tests gave the following results: stock and autogenous house dust, positive; goose feathers, positive; *Penicillium* and *Hormodendrum*, positive. Seven other molds were negative. Tests with numerous inhalants, pollens, and foods were essentially negative, as was caddis fly.

An inhalation test with moths was positive under the following conditions. The patient was instructed to triturate a number of dead webbing clothes moths in a mortar for five minutes. At the end of this time he had a stuffy nose, paroxysms of sneezing, coughing, and on examination, asthmatic râles over the entire chest.

Course.—The patient returned home while a supply of the moths was procured. He had three severe attacks of asthma in a few days. He was later permitted to stay in New York, but in the home of a relative, and was given subcutaneous injections of moth extract once a week, as well as of a combination of *Hormodendrum*, *Penicillium*, and goose feathers once a week. At the same time a continued effort was made to overcome the infestation of his home with moths. Four weeks later he was allowed to go home, and the injections were continued. Until now, nine months after therapy was begun, he has been entirely free from asthma.

ENTOMOLOGIC DATA

The moths were identified as the common or webbing clothes moth (*Tineola biselliella* Hummel) by John W. Cadbury, 3rd, of the Academy of Natural Sciences of Philadelphia, who also provided the following résumé of its life history. This moth is one of the three species which have acquired the habit of living in dwellings and which, in eating, produce the familiar damage to woolen goods, upholstered furniture and furs (Fig. 3). A single female will lay about 100 eggs in one day, or, in cool weather, in a few weeks. Rough-surfaced materials are preferred, and though eggs may be laid on uneatable material like cotton, the young larvae reach food in time. If exposed to full sunlight, eggs die within seventy-five minutes. At temperatures between 68° and 86° F., eggs require four to ten days to hatch.

Larvae, at hatching, measure 0.8 mm. in length, growing to 10.0 mm. They show strong negative phototropism. A feeding tube of the silk spun from the mouth, and of frass and food material is formed. Optimal

conditions for all stages are a temperature of 77° F. and humidity of 75 per cent. Larvae can survive eight and one-half months or more without food, and sometimes the larval stage may last for several years. The pupal stage lasts ten to twenty days, and the familiar adult then emerges. They mate immediately thereafter, and the gravid female is often unable to fly. The male flies more readily, but tends to run, jump, or hide to escape danger. Adults are unable to feed and have no functional digestive system. Breeding is continuous in artificially heated buildings.

Fig. 3.—The common or webbing clothes moth: adult moths above, larvae below. (Courtesy of the Bureau of Entomology and Plant Quarantine of the United States Department of Agriculture.)

COMMENT

It has long been known that asthma and allergic rhinitis may be due to unusual allergens, the discovery of which often depends as much on fortunate circumstances as on the detective ability of the allergist. In this case, when the mother was queried about the presence of molds, she asked, probably associating the words because of the similarity of sound, whether moths might be responsible. Further questioning led to the fact that the house had been infested with moths for years before the patient's birth. The whooping cough probably acted as a predisposing factor to the allergy.

The following points are worthy of brief mention. Since the scratch test with the only moth commercially available, the coddling moth, even in the strongest obtainable concentration, was entirely negative, the etiology of this allergy could easily have been overlooked if we had not also employed the intracutaneous test because of the suggestive history.

The highly positive reaction on scratch testing with the webbing clothes moth suggests that there is a common antigen in both species of moth and probably also a species-specific antigen.

The reason for the failure to react to caddis fly, contrary to the statements of Parlato,[11] is probably due to the fact that caddis flies are rare, if present at all, in New York City, and hence the patient had no opportunity for sensitization.

This case again demonstrates the invaluable aid to be derived from the clinical inhalation test in cases of asthma and coryza of obscure origin, which react favorably to environmental changes.

Finally, this case illustrates the necessity, in cases of strong reaction solely to house dust, of further searching, not only for molds, as is commonly recommended, but for moths as well.

SUMMARY

1. A case is reported of asthma and allergic rhinitis of nine years' duration, due to the common or webbing clothes moth.

2. This allergy was caused by infestation of the patient's home and was entirely relieved on absence from the house.

3. The suggestive history, the positive inhalation and skin tests, and control of symptoms by injections of moth extract justify consideration of this case as one of specific moth allergy. It differs from other cases of moth allergy in the absence of occupational influences.

4. The literature concerning allergic sensitization from the inhalation of air-borne insect emanations is reviewed.

We take pleasure in acknowledging the kind assistance of John W. Cadbury, 3rd, of the Academy of Natural Sciences of Philadelphia, on the entomologic aspects of this paper.

REFERENCES

1. Metcalf, C. L., and Flint, W. P.: Fundamentals of Insect Life, New York, 1932, McGraw-Hill Book Co., p. 183, 260, 257.
2. Parlato, S. J.: Emanations of Flies as Exciting Causes of Allergic Coryza and Asthma: III. Hypersensitiveness to Moths and Butterflies (*Lepidoptera*), J. ALLERGY **3**: 125, 1932.
3. Idem: A Case of Coryza and Asthma Due to Sand Flies (Caddis Flies), J. ALLERGY **1**: 35, 1929.
4. Durham, O. C.: In discussion of Parlato.[2]
5. Balyeat, R. M., Stemen, T. R., and Taft, C. E.: Comparative Pollen, Mold, Butterfly and Moth Emanation Content of the Air, J. ALLERGY **3**: 227, 1932.
6. Parlato, S. J., LaDuca, P. J., and Durham, O. C.: Studies of Hypersensitiveness to the Emanations of the Caddis Fly *(Trichoptera)*: V. A Report of Its Distribution, Ann. Int. Med. **7**: 1420, 1934.
7. Figley, K. D.: Asthma Due to the May Fly, Am. J. M. Sc. **178**: 338, 1929.
8. Caffrey, D. J.: Notes on the Poisonous Urticating Spines of *Hemileuca Oliviae* Larvae, J. Econ. Entomol. **11**: 363, 1918.
9. Randolph, H.: Allergic Response to Dust of Insect Origin, J. A. M. A. **103**: 560, 1934.
10. Wittich, F. W.: The Nature of Various Mill Dust Allergens, Journal-Lancet **60**: 418, 1940.
11. Parlato, S. J.: Emanations of Flies as Exciting Causes of Allergic Coryza and Asthma: V. A Study of the Atopic Reagins of the Caddis Fly, Butterfly, and Moth, J. ALLERGY **3**: 459, 1932.

12. Idem: Studies of Hypersensitiveness to the Emanations of Caddis Flies (*Trichoptera*): IV. Diagnosis and Treatment of Forty-Three Cases of Asthma and Hay Fever, J. A. M. A. **102:** 910, 1934.
13. MacDermot, H. E.: A Case of Asthma Due to May Flies (*Ephemerida*), Canad. M. A. J. **31:** 408, 1934.
14. Parlato, S. J.: The May Fly as an Exciting Cause of Seasonal Allergic Coryza and Asthma, J. ALLERGY **10:** 56, 1938.
15. Figley, K. D.: May Fly (*Ephemerida*) Hypersensitivity, J. ALLERGY **11:** 376, 1940.
16. Unger, L.: In discussion of Figley.[15]
17. Sternberg, L.: A Case of Asthma Caused by *Cimex Lectularis* (Bed Bug), Med. Rec. **129:** 622, 1929; J. ALLERGY **1:** 83, 1929.
18. Ellis, R. V., and Ahrens, H. G.: Hypersensitiveness to Air-Borne Bee Allergen, J. ALLERGY **3:** 247, 1932.
19. Weil, C. K.: Summer Hay Fever of Unknown Origin in the Southeast, J. ALLERGY **11:** 361, 1940.
20. Jamieson, H. C.: The House Fly as a Cause of Allergy, J. ALLERGY **9:** 273, 1938.
21. Kern, R. A.: Asthma Due to Sensitization to a Mushroom Fly (*Aphiochaeta agarici*), J. ALLERGY **9:** 604, 1938.
22. Wittich, F. W.: Allergic Rhinitis and Asthma Due to Sensitization to the Mexican Bean Weevil (*Zabrotes subfasciatus* Boh.), J. ALLERGY **12:** 42, 1940.

Special Reports

HYPERSENSITIVITY TO BEETLES (COLEOPTERA)

Report of a Case

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THE contributions of Figley[1, 2] and of the late Dr. Salvatore Parlato[3, 7] have proved beyond doubt that allergic manifestations can be caused by inhalation of insect emanations. To the group of inhalant insect antigens we herewith add the beetle (*Coleoptera*) with particular reference to the dermestid family.

CASE REPORT

T. D. H., a white male, aged 40 years, whose occupation is that of a museum curator, consulted one of us because of asthma and coryza. He had observed since youth that exposure to excessive house dust would cause mild to moderate asthma. In 1929, soon after accepting the position as assistant curator in a museum, he noticed that his asthma had become more frequent and more severe, and at times it was definitely incapacitating. His duties necessitated the examination of skeletal preparations of various zoological specimens. The skeletons are prepared by allowing dermestid larvae to feed upon the decaying flesh. This feast takes place in a special dermestid room which is carefully closed and shuttered in order to prevent the escape of these insects. The patient had observed that, when he entered the dermestid room, severe asthma promptly ensued. Handling of a freshly prepared skeleton also produced asthma. With the exception of house dust, there was no other inhalant or food allergen suspected to be of etiological importance. Pneumonia at the age of 8 years was the only important previous illness, and the family history revealed that a paternal uncle and one cousin had had asthma. The physical examination, with the exception of an emphysematous chest showing bilateral restriction in motion, and two marble-sized, nontender, freely moveable lymph nodes in the right axilla, was otherwise essentially normal. Urinalysis and blood Kahn test were negative.

Cutaneous tests with protein extracts revealed positive reactions to house dust, to feathers of canary, chicken, and turkey, and to prepared extracts of the adult, larvae, larval skins, and the excretions of the dermestid. Passive transfer tests were repeatedly and strongly positive to the dermestid antigen and mildly positive to house dust. Controls, in adequate numbers, were negative. Additional proof of the specific sensitivity of this patient is that, when he is given an overdose of dermestid extract, or when a powdered preparation of the dermestid is blown into his nose, or when he enters the dermestid room, he develops asthma and rhinorrhea.

A review of the literature on this subject reveals few contributing sources of information. Schwartz and Tulipan[8] mentioned that the bite

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of the beetle will cause an urticarial reaction. Riley and Johannsen[9] briefly mentioned that the *Coleoptera* have been incriminated as a cause of allergic disease. Loir and Legangneux[10] in 1922 reported an occupational disease caused by the *Coleoptera*. The latter authors described a severe, pruritic, erythematous, and vesiculating eruption on the exposed parts of the skin, and a dry, hacking cough in dockmen who were unloading a cargo of bones from a ship found to be heavily infested with dermestid larvae. Powdered material recovered from the hold of this ship was found to be composed of the skins and hairs of the dermestid larvae. Their conclusions were that the larval skins and hairs in powdered form caused an active mechanical irritation of the skin. They also stated that the nasal mucous membranes became moist when the powder was inhaled. Numerous articles have been written regarding the vesicant action of the body fluids and excretion of certain species of beetles. An extract of this type of beetle should not be used because of its powerful vesicant action, the potency of which is not destroyed by heat. The juice from the crushed bodies of these beetles, when rubbed on the skin or on the eyelids, will cause a severe vesiculating dermatitis and conjunctivitis.

The habitat and other entomologic data of *Coleoptera* are adequately covered by Riley and Johannsen,[9] Comstock[11] and Leng.[12]

SUMMARY

1. A case of hypersensitivity to beetle emanations is reported. References to entomologic data of *Coleoptera* are given.

2. The question of whether this source of antigen could be of importance in some cases of environmental allergies is raised.

REFERENCES

1. Figley, K. D.: Asthma Due to the May Fly, Am. J. M. Sc. **178**: 338, 1929.
2. Idem: May Fly (Ephemerida) Hypersensitivity, J. ALLERGY **11**: 376, 1940.
3. Parlato, S. J.: A Case of Coryza and Asthma Due to Sand Flies (Caddis Flies), J. ALLERGY **1**: 35, 1929.
4. Idem: Sand Fly (Caddis Fly) as Exciting Cause of Allergic Coryza and Asthma; Its Relative Frequency, J. ALLERGY **1**: 307, 1930.
5. Idem: Studies of Hypersensitiveness to the Emanations of Caddis Fly (Trichoptera), J. A. M. A. **102**: 910, 1934.
6. Idem: Ibid., Ann. Int. Med. **7**: 1420, 1934.
7. Idem: The May Fly as an Exciting Cause of Seasonal Allergic Coryza and Asthma, J. ALLERGY **10**: 56, 1938.
8. Schwartz, L., and Tulipan, L.: Occupational Diseases of the Skin, Philadelphia, 1939, Lea & Febiger.
9. Riley, W. A., and Johannsen, O. A.: Medical Entomology, New York, London, 1938, McGraw-Hill Book Co.
10. Loir, A., and Legangneux, H.: Accidents de travail occasionnes par des Coleopteres, Bull. Acad. de méd., Paris **88**: 68, 1922.
11. Comstock, J. H.: An Introduction to Entomology, Ithaca, N. Y., 1936, Comstock Publishing Co., Inc.
12. Leng, C. W.: Catalogue of the Coleoptera of America, North of Mexico, Mt. Vernon, N. Y., 1920, John D. Sherman, Jr.

WATER FLEA SENSITIVITY

Case Report

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HEREWITH is reported a case of bronchial asthma caused by sensitivity on inhalation to the water flea (*Cladocera*), a small animal of the phylum *Arthropoda*. It is interesting to note that in contrast with the caddis fly, May fly, butterfly, etc., which are insects, the water flea is a crustacean.

CASE REPORT

F. K., a white male, aged 30 years, had perennial bronchial asthma since the age of 15 years. He had been symptom-free for four years prior to December, 1937, following disposal of his dog. His present illness began on Christmas Eve with symptoms of rhinitis followed the next day by asthma. Symptoms of asthma persisted until the time of taking this history (Jan. 26, 1938). There was no regularity as to their occurrence nor had he any idea of their cause. He kept no pets except fish, and inasmuch as he knew he was sensitive to dog dander, he made a special effort to avoid all animals.

By occupation he was a traveling salesman for a large bakery, and contacted grocers. He spent no time in the bakery and only rarely did he carry samples. He had had fall hay fever since the age of 12 years although he never had an attack of asthma during the season. His mother's mother and his father's father had asthma. The patient had measles, chicken pox, and whooping cough as a child.

His weight was 167 pounds; prior to Christmas, 1937, he weighed 190 pounds. Except for the presence of the typical symptoms and signs of a mild attack of asthma, physical examination revealed nothing unusual. Scratch tests to the usual inhalant materials and foods gave immediate positive reactions to the ragweeds only. Intradermal tests gave small immediate reactions to a number of other inhalents with larger reactions (noted as two plus) to dusts. The laboratory tests, including urinalysis, blood counts, and serologic tests for syphilis, revealed nothing of significance.

The patient was advised to observe the usual precautions regarding house dust and orris root. He was supplied with a nebulizer for the use of epinephrine 1:100 by inhalation, and treatment was started with hypodermic injections of extracts of house dust and ragweed pollen. No improvement could be shown except for the relief obtained with the nebulizer. In fact, he experienced three acute attacks of asthma which lasted for several days and which occurred about the middle of three successive months (March, April, and May, 1938). During the course of repeated and persistent questioning, he casually mentioned having gone to a dealer to buy some supplies for his tropical fish which he raised as a hobby. It then developed that he had several large tanks of fish in his home and not a small bowl as was at first presumed. Following that lead, it was learned that he had been in the habit of going to the dealer's store about every month, while further inquiry elicited the information that now the matter had been brought to his attention, he felt quite certain he had noticed something about those visits which did not agree with him.

Read in part before the Cleveland Allergy Society, Jan. 30, 1940.

His purchases consisted chiefly of three varieties of fish food, two of which he said contained *Daphnia* (one of the water fleas). Scratch tests with each of these materials gave large wheals to those containing *Daphnia*. Passive transfer studies were done on sites prepared in two nonallergic individuals and tested with the *Daphnia* extract by the scratch method twenty-four hours later. Wheals varying from 0.5 to 1 cm. in diameter appeared quite promptly while the control areas were negative. Intradermal tests on the same sites one week later produced wheals 2 cm. in diameter, the control areas showing no reaction. Conjunctival and nasal tests with the dried material produced symptoms typical of an allergic reaction. The inhalation of this material was followed by an attack of bronchial asthma within five minutes.

The patient was advised to discontinue the use of *Daphnia* following the appearance of his positive scratch test to the material. He has had no acute attacks of asthma since that time. This report covers a period of observation extending for nearly three years after the discovery of the offending agent.

Treatment with ragweed pollen extract given hypodermically was continued through the fall of 1938 with fair results. He experienced but seventeen days of mild hay fever.

Water fleas "are abundant everywhere in fresh water. One of the commonest species, *Daphnia pulex*, found in ponds and ditches, is less than 0.1 inch in length.* The female carries the eggs in a brood-chamber between the back of the body and the shell until hatching takes place. Throughout the greater part of the year only females occur and the eggs develop . . . without fertilization."[2]

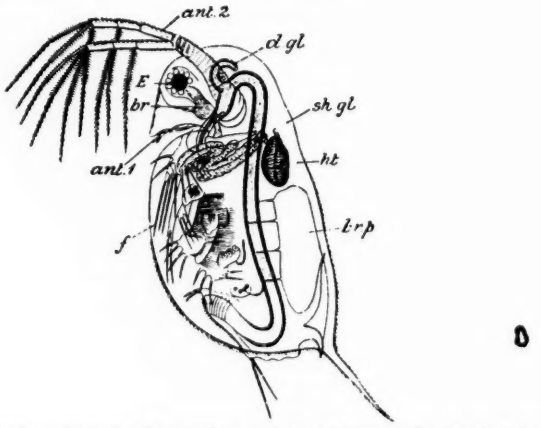

Fig. 1.—*Daphnia. Ant. 1*, antennule; *ant. 2*, antenna; *br*, brain; *br.p*, brood pouch; *d.gl*, digestive gland; *E*, eye; *f*, swimming-feet; *ht*, heart; *sh.gl*, shell-gland. The small figure to the right represents the actual size of the dried specimen. (Photograph by Fred T. Locke by permission of The Macmillan Co.)

Water fleas are "important food for fishes. Their importance in the economy of water is largely due to their very rapid rate of reproduction. During the summer season broods of eggs successively appear at intervals of only a few days. The young develop rapidly and are themselves soon producing eggs. In *Daphnia pulex*, for example, it has been calculated that the possible progeny of a single female might reach the astounding number of 13 billion in 60 days."[3]

*For detailed zoologic data see references 1, 2, and 3.

There are a number of species of water fleas which resemble one another so closely that positive identification requires expert determination. The fish food referred to in this report was labeled as *Daphnia* and has been definitely identified as being one of the water fleas (*Cladocera*) and very likely of the genus *Daphnia*. The condition of the dried material would not permit naming the species.[4]

Fig. 2.—Showing the appearance of the broken-up, dried water fleas (×250). (Photograph by the late Edward L. Saylor, M.D.)

Even with careful handling the material readily disintegrates to form a dust. It is believed that it was the inhalation of this dust in handling the material in the feeding of fish which provoked the attacks of acute bronchial asthma in the instance just cited.

SUMMARY

A case of bronchial asthma caused by sensitivity on inhalation of the water flea is reported.

Indirectly, fish may be placed with other animals as responsible for inhalant sensitivities in allergic persons.

REFERENCES

1. Parker, T. Jeffery, and Haswell, William A.: A Textbook of Zoology, London, 1921, The Macmillan Company, vol. 1, p. 558.
2. Calman, William Thomas: Water flea, in The Encyclopediae Britannica, ed. 14, London, 1937, Encyclopediae Britannica Co., Ltd., vol. 23, p. 413.
3. Needham, James G., and Lloyd, J. T.: The Life of Inland Waters, Ithaca, N. Y., 1916, Comstock Publishing Company, pp. 185-7.
4. Kraatz, Walter C., Professor of Zoology, University of Akron: Personal communication.

COCOBOLO WOOD DERMATITIS

Report of a Case Due to Wooden Handles on Kitchen Knives

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CONTACT dermatitis caused by woods is a frequently recognized condition. Senear[1] has set forth in great detail the many woods which have been implicated in such cases. One of these, cocobolo wood, has been the subject of prior reports, but despite the fact that much of the output of this wood is used for the manufacture of knife handles, no previous case due to this type of contact can be found in the literature.

Cocobolo wood is produced in the tropics and is of three chief species *Dalbergia retusa* (nambar wood); *Dalbergia hypoleuca* (palo negro nambar); *Dalbergia granadillo* (Costa Rica rosewood). It is a beautiful, hard, oily or resinous wood, varying in color from light yellow to deep mahogany. It is sometimes called "bastard mahogany." It comes from trees indigenous to the West Indies, Mexico, and South America.

About three-fourths of the supply of cocobolo wood is used by manufacturers of cutlery for handles of table and carving knives and forks. It is also used for the handles of pocket knives and butcher knives. The oily nature of the wood tends to waterproof it and a high natural finish results from merely rubbing it. This wood is also used for turned articles such as small tool handles, chessmen, canes, jewelry boxes, wooden bracelets, wooden cut-out brooches, knobs on cabinet doors, policemen's clubs, and many novelties. It is frequently employed in the manufacture of fancy brushes and mirrors where there is a demand for a wood of beautiful grain and color.

The first patients with dermatitis due to cocobolo wood were reported by Stern in 1891.[2] These were both flute players who developed a contact dermatitis of the lips from the mouthpiece of the instrument. MacKee[3] reported a case in a man whose occupation was scraping the backs of hairbrushes prior to their being stained and varnished. He had, however, no contact with the latter items. Schröpl, in 1935,[4] reported a case in an instrument maker who was exposed to cocobolo wood dust coming from an adjoining room. This produced a very severe dermatitis on the exposed surfaces of his body. A case due to this wood was reported by Abramowitz and Swarts.[5] Their patient developed the dermatitis while carving a knitting-bag

handle of cocobolo wood. Levin and Behrman,[6] in 1939, reported a case due to a cocobolo wooden bracelet. The following case due to the wooden handles of kitchen knives is apparently the first report of such an occurrence.

CASE REPORT

Miss E. L. H., aged 51 years, was employed as a court stenographer. Her family history was negative for allergy. She had had no previous allergic difficulties. She recalled, however, that her wrist had become irritated several years ago when she had worn a wooden bracelet. Her present difficulty began in December, 1939.

Fig. 1.

Fig. 2.

Figs. 1 and 2.—Photographs of patient's hands on May 10, 1940, showing marked erythema, induration, desquamation, and fissuring. Note serous exudate on the fingers and palms of the hands and edema of the fingers.

At that time she noticed that her hands and fingers began to itch, and the skin appeared to be darker in color. There was a gradual thickening of the skin, accompanying by scaling and fissuring. The lesions spread over the back of the hands and fingers, which began to swell to such an extent that she found it difficult to continue her work. A few months after the onset she noticed a similar involvement of her face and neck, and she observed that she was unable to continue the use of her previously well-tolerated cosmetics, since these items seemed to aggravate the facial condition. There was no history of a change in her occu-

pation or environmental factors except that she had moved into a new apartment a few months before the onset of her present illness. She had consulted a number of physicians and had received a great deal of local treatment which apparently had been directed chiefly against a suspected fungus infection. She had also received a great many x-ray treatments, without benefit. In the spring of 1940 she spent a two-week vacation in Florida, during which her skin cleared up almost completely. The condition recurred, however, within a week after returning to Detroit.

When she was first seen on March 13, 1940, she presented a very severe eczematous dermatitis of both hands, extending as far as the wrists. There was marked scaling accompanied by vesiculation and thickening of the skin on the palms of the hands and marked edema of the fingers. There were numerous fissures in the skin. Movement of the fingers was painful.

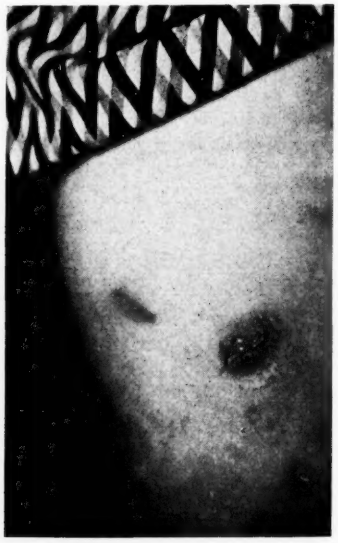

Fig. 3.—Patch tests from a sliver removed from the patient's large handled kitchen knife and from scrapings of her small handled knife.

The possibility of a contact dermatitis due to wood was suggested because of the history of irritation several years before from a wooden bracelet. Patch tests were made for all the woods with which she came in contact, including the wooden handles on her kitchen knives. Patch tests were also made for many other contactants, including cosmetics, articles of clothing, and many items used in her work. All were negative except those made from the wooden handles of the kitchen knives. These produced very strongly positive skin reactions with severe vesiculation requiring several weeks to heal. The patient was asked to discard the kitchen knives; improvement began immediately and continued rapidly, so that in six weeks' time the skin was entirely clear.

An ether extract was prepared from fresh cocobolo wood shavings supplied by the manufacturer. One drop of this was applied to the skin of the patient's thigh. She was instructed to remove this with ether or benzene at the first sign of irritation, but she overlooked this precaution. When seen twenty-four hours later, she had developed very severe vesiculation of the skin, with subsequent ulceration requiring several weeks to heal. A number of other allergic patients who were patch tested with this extract gave negative tests.

In a personal communication from one of the factories making cocobolo wood handles for cutlery, it was stated that workmen not infrequently developed a dermatitis resembling poison ivy from the dust produced in the working of the wood.

SUMMARY

About three-quarters of the entire output of cocobolo wood is used for cutlery handles.

A case of severe contact dermatitis due to cocobolo wood handles on kitchen knives is reported.

The possibility of sensitization to this substance in cases of dermatitis of the hands should be kept in mind.

REFERENCES

1. Senear, F. E.: Dermatitis Due to Woods, J. A. M. A. **101**: 1527, 1933.
2. Stern, E.: Ueber einige Hautkrankheiten der Musicker: ein Beitrag zu den professionellen Dermatosen, München. med. Wchnschr. **38**: 739, 1891.
3. MacKee, G. M.: Dermatitis Venenata From Cocobolo Wood, J. Cutan. Dis. **31**: 582, 1913.
4. Schröpl, E.: Eczema From Granadilla Wood, Urol. & Cutan. Rev. **39**: 571, 1935.
5. Abramowitz, E. W., and Swarts, W. B.: Dermatitis Due to Cocobolo Wood, Arch. Dermat. & Syph. **37**: 441, 1938.
6. Levin, O. L., and Behrman, H. T.: Bracelet Dermatitis, New York State J. Med. **39**: 1877, 1939.

469 Fisher Building

THE LINDEN (TILIA) AS A FACTOR IN SEASONAL POLLINOSIS

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OCCASIONALLY one finds a plant which conforms in all respects to Thommen's postulates and which might therefore cause hay fever or asthma, and yet has been overlooked as an etiological agent in seasonal pollinosis. We believe this to be true of the linden, *Tilia*. We have been able to find no other cases of linden sensitization, and there is no mention of it in any of the standard texts on allergy.

In the four cases to be described, positive reactions were obtained to extracts of linden pollen by the scratch technique, by the conjunctival test, and by passive transfer. In addition, the symptoms coincided with the time of pollination, and two of the patients lived in close proximity to large numbers of linden trees. Two other positive skin reactions to linden pollen extract were encountered, but here there was not a definite correlation between the time of symptoms and of pollination; hence these are not included.

Surveys to determine the incidence of tree pollinosis have been made by a number of authors. Vander Veer, Cooke, and Spain,[1] in a series of 2,836 cases of pollinosis, found that cases of pure tree pollinosis comprised less than 1 per cent of their material while cases of mixed pollinosis (trees plus other pollens) totalled 8 per cent. Bernton,[2] in reviewing 791 cases of hay fever, found that 4.2 per cent of the patients were sensitized clinically solely to trees. Tuft and Blumstein[3] stated that cases of mixed pollinosis accounted for 6.5 per cent of 108 cases of hay fever. Vaughan[4] found that his cases of tree pollinosis ranged from 1 to 8 per cent. On the whole, therefore, trees play a part in approximately 5 per cent of the cases of pollinosis. There seems to be a rather general agreement, among allergists east of the Mississippi, that the important trees are oak, hickory, sycamore, poplar, maple, elm, and ash. It is proposed that linden be added to this list.

CASE REPORTS

CASE 1.—G. D., a white male aged 6 years, who lived on a boulevard lined with lindens, had hay fever from May to June (linden) and again from mid-August until frost (ragweed). His skin test was positive by scratch test to linden pollen, and the passive transfer test was positive. It is interesting to note that when the ophthalmic test was performed, enough of the pollen was carried into the nose through the nasolacrimal duct to produce a positive nasal contact test, with rhinorrhea, in addition to the positive conjunctival test. His mother has spring hay fever and urticaria due to foods.

From the Vaughan-Graham Clinic.
*Upjohn Research Fellow.

CASE 2.—A. G., a white female aged 33 years, has had hay fever for the past 10 years, starting in May and June, and clearing up in July, to return again in the fall. She has asthma in the spring. She gave positive skin reactions to house dust, ragweed, and linden pollens, and positive ophthalmic and passive transfer tests to linden. No familial history of allergic disorders was noted.

CASE 3.—W. T., a white male aged 36 years, has had hay fever and asthma since 1930. His hay fever always occurs from early May to mid-June, lasting about six weeks. He gave skin reactions to trees only, including the following: black walnut, pecan, hickory, cottonwood, yellow birch, oak, ash, and linden. The conjunctival and passive transfer tests were positive to linden. His father has asthma.

CASE 4.—M. S., a white male aged 26 years, living in a grove of lindens has had asthma since he was 9 years old. His asthma is seasonal, occurring from mid-February to the latter part of September. He has no nasal symptoms at all. He reacted to redtop, hackberry, short and giant ragweed, and linden. Ophthalmic and passive transfer tests were positive to linden.

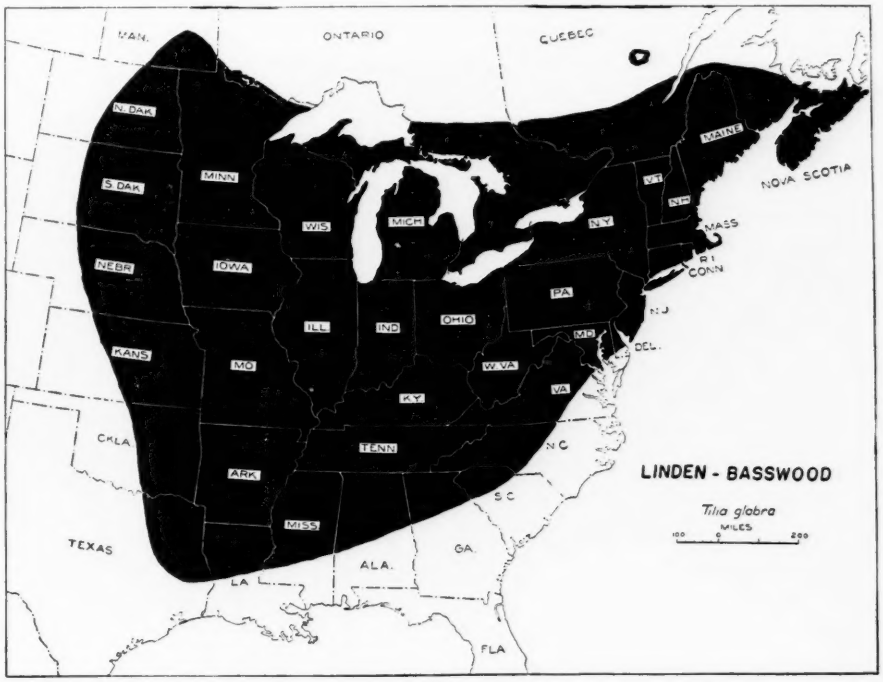

Fig. 1.—Distribution of linden trees. (Courtesy of the Forest Service, U. S. Dept. of Agriculture.[11])

The four cases have been found in a period of three months only. Hence linden sensitization may be fairly common in areas where linden trees are relatively abundant.

DISCUSSION

According to Wodehouse,[5] the family *Tiliaceae* comprises a large number of trees, shrubs, or rare herbs, represented in the northern regions by the single genus, *Tilia*. This is a native of the rich woods in the

northern states and Canada and reaches its greatest size in the valley of the lower Ohio, becoming 130 feet in height; its usual height is about 70 feet. The trunk is erect and pillarlike; the branches are spreading, pendulous, forming a broad, rounded head (Keeler[6]). Britton and Brown[7] list four species as occurring in the northern United States: *T. americana, T. pubescens, T. heterophylla,* and *T. michauxii.*

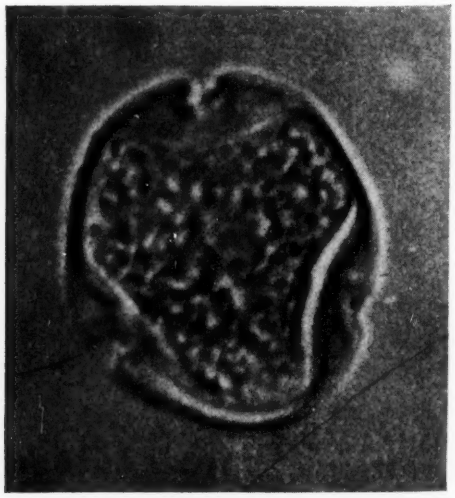

Fig. 2.—Photomicrograph of linden pollen in oil (high dry objective).

The following description of the pollen is from Wodehouse.[5]

"*Tilia americana* pollen grains are uniform in size and shape, oblately flattened, and about 36.5 × 28 μ; germinal pores three, rarely two or four, deeply sunken, elliptical in shape, and equally spaced on the equator. The extine is reticulate pitted. Furrows in the ordinary sense of the word are absent, but the pits in which the pores are situated are slightly elongate in a meridional direction and must undoubtedly be regarded as vestigial furrows. Underlying each pore is a relatively large thickening of the intine which forms a very conspicuous feature of the grain in optical section and causes the region surrounding the pore to be elevated slightly. It flowers [in the northern states] in May and June. It is insect pollinated but generally sheds much pollen. It is not a factor in hay fever but its pollen is frequently caught on atmospheric pollen plates."

Since the pollen is primarily entomophilous, one would expect sensitization to occur where there is a large number of lindens. The distribution in Richmond consists of scattered trees throughout yards and one linden-lined boulevard, said to be the longest of its kind with the exception of the world-famed Unter den Linden in Berlin.

In his text on allergy (1935), Urbach[8] states that he is of the opinion that in the so-called "Lindenheuschnupfen" one is dealing, for the most part, with an idiosyncrasy not conditioned by linden pollen but through

the scent (Duftstoffe) and probably the ethereal oil. He mentions that he has seen persons who were allergic to grass pollens in May, but who at the end of June were allergic only to the aroma of linden blossoms. He bases this statement on the fact that a nasal test with linden pollen gave negative results, whereas the inhalation of a bouquet of linden blossoms immediately produced a sneezing attack and rhinorrhea. He thought he was dealing with a hypersensitivity to the ethereal oils in the blossoms. In another communication, Urbach and Wiethe[9] emphasized that hay fever sufferers may have symptoms from the aroma of strongly odorous plants such as jasmine, acacia, or linden. They attribute this to the ethereal oil (aetherische Oele) by which they designate those products which are derived from plants by moist distillation and which produce the characteristic odor of the plant. This oil consists of several components, some unknown, but many products of terpene ($C_{10}H_{16}$) are present and also nitrogen-containing substances, e.g., anthranil acid esters, indol, skatol, etc.

Urbach,[8] in his text on allergy, cited a case of eczema due to contact with linden blossoms or leaves, but examination of the original article has failed to confirm this.*

COMMENT

The proofs of the specificity of the linden pollen in the causation of the symptoms in our cases are: the synchronism of symptoms and pollination; the fact that the pollen is partially anemophilous; the demonstration of skin and mucous membrane sensitivity to the pollen; and finally, the demonstration of the presence of circulating skin sensitizing reagins (Prausnitz-Küstner antibodies) by passive transfer tests.

SUMMARY

Four cases of seasonal pollinosis due to the linden *(Tilia)* are reported.

It is suggested that linden be added to the list of pollens with which patients may be routinely tested.

REFERENCES

1. Vander Veer, A., Cooke, R. A., and Spain, W. C.: Diagnosis and Treatment of Seasonal Hay Fever, Am. J. M. Sc. **174:** 101, 1927.
2. Bernton, H. S.: Tree Pollinosis, Virginia Med. Monthly **57:** 170, 1930.
3. Tuft, Louis, and Blumstein, George: Incidence and Importance of Tree Pollen Hay Fever, With Particular Reference to Philadelphia and Vicinity, J. ALLERGY **8:** 464, 1937.
4. Vaughan, Warren T.: Allergy and Applied Immunology, ed. 2, St. Louis, 1934, C. V. Mosby Co.
5. Wodehouse, R. P.: Pollen Grains, New York and London, 1935, McGraw-Hill Book Co., p. 442.
6. Keeler, Harriet L.: Our Native Trees, New York, 1927, Chas. Scribner's Sons.

*Although Kletetschka[10] described an eczema caused by a linden, it was caused by the "Zimmerlinde" or room linden (*Sparmannia africana*), a potted plant unrelated to the linden (*Tilia*).

7. Britton, N. L., and Brown, Addison: An Illustrated Flora of the Northern United States, Canada, and the British Possessions, New York, 1913, Chas. Scribner's Sons.
8. Urbach, Erich: Klinik und Therapie der Allergischen Krankheiten, Vienna, 1935, Maudrich, p. 101.
9. Urbach, Erich, and Wiethe, C.: Aetherische Oele als Ursache von allergischen Haut- und Schleimhauterkrankungen, München. med. Wchnschr. **78:** 2030, 1931.
10. Kletetschka, A.: Erworbene Pflanzen—überempfindlichkeit, Dermat. Wchnschr. **96:** 86, 1934.
11. Munns, E. N.: The Distribution of Important Forest Trees of the United States, Washington, April, 1938, U. S. Dept. of Agriculture, Miscellaneous Publication No. 287.

HYPERSENSITIVITY TO THIAMIN CHLORIDE, WITH A NOTE ON SENSITIVITY TO PYRIDOXINE HYDROCHLORIDE

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WHILE it is generally believed that thiamin chloride may be administered without the risk of harmful effects,[1] a few writers have warned against possible dangers. Ochsner and Smith[2] and Stern[3] mentioned the lethal effects produced in animals by large experimental doses. Steinberg[4] reported two patients in whom symptoms developed after long-continued use of thiamin chloride. He felt that the reactions encountered in these cases might be due either to a sensitivity developing toward vitamin B_1 or to supersaturation of the tissues of the body with it.

The following two cases are reported to emphasize the possibility that prolonged thiamin chloride administration may result in the development of hypersensitivity.

CASE 1.—Mrs. A. B. was 59 years old. Thiamin chloride was first administered in August, 1937, and was taken intermittently by mouth throughout the next two years. Beginning in December, 1939, thiamin chloride was given by hypodermic injection, fairly regularly and usually twice weekly, in doses ranging from 8 to 90 mg. Liver extract was usually given in conjunction with the thiamin, and at times other factors of the B complex were included in the injection. An oral polyvitamin preparation was also taken throughout this period.

On May 17, 1940, a few minutes after the injection of 30 mg. of thiamin chloride combined with liver extract and a crude B complex preparation, transient sensations of weakness and faintness developed.

On May 21, 1940, following a similar injection the patient complained of excessive nervousness. The pulse rate rose from 70 to 120, and the face and hands were flushed. The symptoms subsided in less than one-half hour. The possibility of a nicotinic acid reaction was considered, and no further injections were given for a week.

On May 28 an injection of liver extract was followed by no reaction.

On June 1 injection of liver extract combined with 30 mg. of thiamin chloride was followed by a reaction similar to, although more severe than, that encountered on May 21, suggesting that thiamin, rather than nicotinic acid, was the responsible agent.

During the following two weeks the patient several times attempted to take polyvitamin capsules and capsules containing liver extract, iron, and thiamin chloride. In each instance nervousness, associated with some flushing and tachycardia, developed within an hour following ingestion of the capsules.

On June 25 an intracutaneous injection of 3 mg. of thiamin chloride was followed by the development of a wheal approximately 2.5 cm. in diameter. At the

same time the patient became very nervous; the pulse rose to 140; and the face was flushed. Later there was profuse perspiration. The symptoms subsided in about thirty minutes.

Subsequently the intracutaneous injection of 0.15 mg. of thiamin in 0.03 c.c. of salt solution produced an irregular wheal measuring 2.0 by 1.9 cm., with a surrounding zone of erythema measuring 5 by 5 cm. in its greatest diameters. Similar injections in five control subjects produced no local or general reaction although the intracutaneous injection of larger quantities of thiamin, 2.5 mg. in 0.5 c.c. of fluid, did result in wheal formation.

An intracutaneous injection of chlorbutanol, the preservative used in the thiamin chloride solution, gave no local or general reaction. The administration of nicotinic acid by mouth, in doses up to 150 mg. daily, produced no reaction.

Several months later the oral administration of thiamin was begun, the initial dose being 0.015 mg. The size of the dose was gradually increased, until, at the time of writing, 0.3 mg. was being taken three times daily, with no apparent reaction.

CASE 2.—Mrs. C. D., aged 56 years, for several years took thiamin chloride intermittently, both by mouth and by injection. During the summer of 1940 several injections were given at intervals of three or four days in doses of 20 to 50 mg. An hour or more after each of the last few of these injections a sensation of heat developed, followed rather quickly by profuse perspiration and by marked tinnitus. As these symptoms subsided feelings of agitation and panic developed, lasting up to twenty-four hours. Oral administration of thiamin chloride produced similar, but milder, symptoms.

An intracutaneous test dose of 3 mg. of thiamin chloride produced a wheal about 2.0 cm. in diameter, although the usual sequence of general symptoms did not follow. An intracutaneous injection of chlorbutanol produced no reaction.

After discontinuing the use of thiamin, the patient was given a series of semiweekly injections of pyridoxine hydrochloride in doses up to 25 mg. While the response at first was entirely favorable, after about two months symptoms began to develop similar to those which had followed the thiamin chloride injections. Intracutaneous injection of 2.5 mg. of pyridoxine at this time produced a wheal about 3.5 cm. in diameter. A similar injection in patient A. B. produced no local or general reaction. Similar injections in three control subjects produced small wheals, in no case greater than 1.1 cm. in diameter.

SUMMARY

1. Two cases are reported in which the prolonged administration of thiamin chloride resulted in the production of severe symptoms, apparently due to the development of hypersensitivity.

2. Intracutaneous injection of 0.15 mg. of thiamin resulted in definite wheal formation, although similar injections produced no reaction in control subjects.

3. Subsequently small quantities of thiamin were safely administered by mouth to one patient.

4. The repeated injection of pyridoxine hydrochloride in the second patient was followed after about two months by the development of symptoms similar to those which had followed thiamin injection.

5. Intracutaneous injection of pyridoxine resulted in definite wheal formation in this patient, although it produced no reaction in the first patient, and only questionable wheal formation in three control subjects.

ADDENDUM

Since the preparation of this paper a communication from Mills[5] has appeared stressing possible toxic reactions to thiamin. Cases were cited in which the administration of thiamin was followed by symptoms similar to those encountered in the cases reported here. In Mills' cases apparently the symptoms developed gradually following prolonged oral administration, rather than immediately following injection.

REFERENCES

1. (a) Moore, M. T.: Treatment of Multiple Sclerosis With Nicotinic Acid and Vitamin B_1. Preliminary Report, Arch. Int. Med. **65**: 1, 1940.
   (b) Metildi, P. F.: The Treatment of Tabetic Lightning Pains With Thiamin Chloride, Am. J. Syph., Gonor., & Ven. Dis. **23**: 1, 1939.
   (c) Meiklejohn, A. P.: The Water-Soluble Vitamins, New England J. Med. **220**: 518, 1939.
2. Ochsner, A., and Smith, M. C.: The Use of Vitamin B_1 for the Relief of Pain in Varicose Ulcers, J. A. M. A. **114**: 947, 1940.
3. Stern, E. L.: The Intraspinal Injection of Vitamin B_1 for the Relief of Intractable Pain and for Inflammatory and Degenerative Diseases of the Central Nervous System, Am. J. Surg. **39**: 495, 1938.
4. Steinberg, C. L.: Untoward Effects Resulting From the Use of Large Doses of Vitamin B_1, Am. J. Digest. Dis. **5**: 680, 1938.
5. Mills, C. A.: Correspondence, J. A. M. A. **116**: 2101, 1941.

Society Proceedings

FIRST ANNUAL REPORT OF THE ADVISORY BOARD OF AMERICAN ALLERGISTS, MAY, 1941

Following action taken at their annual meetings in 1940 by the Society for the Study of Asthma and Allied Conditions and the American Association for the Study of Allergy, the Committee on Standards, a joint committee of the two societies, became the Advisory Board of American Allergists. The members of the superseded Standards Committee were designated by the respective presidents to this Board.

During the past year the Board has considered or carried out the following:

1. It has entered into an agreement with the American Board of Internal Medicine whereby the Advisory Board of American Allergists is the recognized body for all allergy groups to deal with the question of examination and certification of specialists in Allergy, following a candidate's certification as "Internist" by the American Board of Internal Medicine. This agreement has been formally accepted by the Advisory Board for Medical Specialties and the Council on Medical Education and Hospitals of the American Medical Association. This agreement is identical with that of the other cooperating subspecialty groups in cardiovascular disease, gastroenterology, and tuberculosis.

2. Following the recommendation of your Board, the American Board of Internal Medicine has certified as specialists in Allergy all those members of the two national allergy societies who have already received certification as "Internists." Such certificates are now being issued.

3. This Advisory Board has prepared a list of guest examiners to act with the American Board of Internal Medicine in examinations for certification in the subspecialty Allergy. One such examination was held in Boston in April. Dr. Francis Rackemann was guest examiner. One candidate was examined and recommended for certification. Dr. Milton Cohen and Dr. Warren Vaughan will be guest examiners in Cleveland in June.

4. A number of allergy clinics have submitted records to this Board during the past year. A list of those approved by the Board is appended to this report. Your Board feels that this method of examination for approval is far from ideal and has discussed ways and means of conducting personal inspections, but no action toward this end is feasible at this time. Records submitted are often unsatisfactory because of a meager history, lack of complete physical examination and laboratory

work-up. The skin tests are often extensive but pointless, and there is no adequate discussion and diagnosis. Clinics applying for "approval" should pay greater heed to these points in their clinic records.

5. The Advisory Board has cooperated with the American College of Physicians regarding post-graduate courses in Allergy. Two such courses have been given this year, one at the Massachusetts General Hospital and one at the Roosevelt Hospital. As there is a demand for these short courses for general practitioners and internists, steps have been taken to increase the number of allergy clinics participating in this work.

6. A Committee on Graduate and Post-Graduate Education in Allergy, representing both national societies, was appointed, with Dr. Matthew Walzer, chairman. This Committee has attempted a survey of the existing faculties for such education. It has reported to this Board the information received. It is unfortunate that only 65 per cent of clinics already approved had supplied data to the Committee. The Committee was requested to continue its efforts. This is a time-consuming piece of work and should be heartily supported by all approved clinics.

7. A report of the Committee on Workmen's Compensation of the Society for the Study of Asthma and Allied Conditions was referred to this Board. No action was taken other than to send a copy to Dr. Harry Foerster, chairman of a similar committee of the American Medical Association, for comment and advice. No reply has yet been received.

Respectfully submitted by the Committee

ROBERT A. COOKE
RICHARD A. KERN
J. ALEXANDER CLARKE
LESLIE N. GAY
GEORGE PINESS
FRANCIS M. RACKEMANN

Hospitals With Allergy Clinics Approved by the Advisory Board of American Allergists, Representing the American Society for the Study of Allergy, and the Society for the Study of Asthma and Allied Conditions, up to June 1, 1941

ARIZONA

Phoenix

1. St. Luke's Home

CALIFORNIA

Los Angeles

2. Children's Hospital
3. Los Angeles County General Hospital
4. Santa Fe Coast Lines Hospital

DISTRICT OF COLUMBIA

Washington

5. Children's Hospital

INDIANA

Indianapolis

6. James Whitcomb Riley Hospital for Children

MARYLAND

Baltimore

7. Johns Hopkins Hospital
8. University Hospital

MASSACHUSETTS

Boston

9. Beth Israel Hospital
10. Massachusetts General Hospital

MICHIGAN

Ann Arbor

11. University Hospital of the University of Michigan

MINNESOTA

Rochester

12. Mayo Clinic

MISSOURI

St. Louis

13. Barnes Hospital
14. Firmin Desloge Hospital
15. McMillan Hospital

NEW JERSEY

Montclair

16. Mountainside Hospital

NEW YORK

Brooklyn

17. Beth-El Hospital
18. Jewish Hospital

Buffalo

19. Buffalo General Hospital
20. Children's Hospital of Buffalo

New York

21. Bellevue Hospital, Third Division
22. Bellevue Hospital, Children's Division
23. Beth-Israel Hospital
24. French Hospital
25. Gouverneur Hospital
26. Lebanon Hospital
27. Midtown Hospital
28. Mount Sinai Hospital
29. New York Hospital
30. New York Eye and Ear Hospital
31. New York Infirmary for Women and Children
32. New York Polyclinic Medical School and Hospital
33. New York Post-Graduate Hospital, Department of Medicine
34. New York Post-Graduate Hospital, Department of Pediatrics
35. New York Skin and Cancer Unit, New York Post-Graduate Medical School and Hospital
36. New York University Medical Clinic
37. Roosevelt Hospital
38. St. Luke's Hospital
39. Sydenham Hospital

Rochester

40. Rochester General Hospital
41. Strong Memorial Hospital

Syracuse

42. Syracuse Free Dispensary

PENNSYLVANIA

Philadelphia

43. Children's Hospital
44. Germantown Hospital
45. Graduate Hospital of the University of Pennsylvania
46. Hospital of the University of Pennsylvania
47. Jefferson Hospital
48. Mount Sinai Hospital
49. Pennsylvania Hospital
50. Philadelphia General Hospital
51. Temple University Hospital

Pittsburgh

52. University of Pittsburgh Hospital, Falk Clinic
53. United States Veterans Administration Hospital

RHODE ISLAND

Providence

54. Rhode Island Hospital

VIRGINIA

Richmond

55. Medical College of Virginia, Hospital Division

Selected Abstracts

Asthma and Hay Fever

UNDER THE DIRECTION OF SAMUEL M. FEINBERG, M.D., AND THEODORE B. BERNSTEIN, M.D., CHICAGO

Observations on the Effect of Injection of Pollen Extracts on the Skin Sensitivity of Hay Fever Subjects. Bruce Pearson, R. S.: Guy's Hosp. Rep. **20**: 55, 1940.

Over a period of three years the author observed the effect of treatment upon the skin reaction in a group of English subjects whose hay fever was due to grass pollen. Both scratch tests and intradermal tests with serial dilutions of three grass pollen extracts, meadow grass, fox tail, and Yorkshire fog were made. The tests were done each year before institution of treatment, at the end of treatment, and three to four months after treatment. The group, in general, showed a slight diminution of skin sensitivity immediately after treatment as revealed by the intradermal test. Often an extract ten or 100 times more concentrated was necessary to elicit a minimal reaction after treatment. This diminution in skin reactivity was temporary, since skin sensitivity nearly always returned to normal, as shown by the results of testing three to four months after treatment. The scratch test after treatment almost always showed a marked diminution in the size of its response, and sometimes became negative temporarily, only to return to its original size three to four months later. The temporary desensitization that occurred was no greater after three years of treatment than after one. It was found that local desensitization was more marked at the site of the weekly injections than elsewhere. The author found no evidence to support the view that injection of individual grass pollen extracts leads to specific desensitization to the pollens in question and not to other species of grass pollen.

Studies With Antigens. VII. The Significance of Reactions to Intracutaneous Tests Performed With Extracts of Purified House Dust. Efron, B. G., Boatner, C. H., and Pabst, M. A.: J. Invest. Dermat. **4**: 95, 1941.

The authors performed intracutaneous tests with purified house dust extract in dilutions ranging from 0.002 to 0.000002 per cent in a control group of sixty-seven persons with no allergic disease and a group of seventy-three subjects who had perennial hay fever or asthma, or both. The incidence of positive reactions in the control group to concentrations of house dust extract of 0.002 per cent or less was not significantly different from the incidence of reactions to scratch tests made with 0.87, 0.75, and 0.50 per cent solutions of the same material. The incidence of these reactions had been shown previously to be 7.00 ± 1.80 per cent to 8.50 ± 1.97 per cent and is of no concurrent clinical significance. Likewise, in the allergic group, the incidence of reactions obtained by the intracutaneous method with dilutions of 0.002 to 0.0002 per cent did not significantly differ from the incidence of positive reactions obtained by the scratch method with 0.87, 0.75, and 0.50 per cent solutions. The incidence of positive scratch reactions in this group had previously been shown to be 75.62 ± 3.39 per cent to 78.75 ± 3.24 per cent. The number of positive reactions obtained in this group when tested intracutaneously with solutions of 0.0001 to 0.000002 per cent was significantly less than the number of reactions obtained by scratch tests with house dust solutions of 0.87, 0.75, and 0.50 per cent. These

extracts used intracutaneously up to a dilution of 0.002 per cent are not irritating, and the reactions produced by them are allergic in nature.

Immunological Studies of Pollinosis. II. Passive Sensitization of Man Through Transfusion. Loveless, M. H.: J. Immunol. **41**: 15, 1941.

Three nonallergic patients, hospitalized because of Banti's disease, lymphosarcoma, and Stell's disease, respectively, were transfused with reagin-containing blood from five persons who had ragweed pollinosis. These ragweed-sensitive people had not received any hay fever treatment prior to the use of their blood. The three recipients were proved to be not allergic to ragweed pollen by skin, conjunctival, and passive transfer tests.

After transfusion each of the three nonsensitive subjects exhibited the findings of passive sensitization. The sensitizing factor in the transfused blood was taken up by the skin, conjunctiva, and nasal mucosa; these findings were demonstrated by appropriate testing. The skin was the first tissue to show a reaction to ragweed extract and the last to lose sensitivity. However, considerable variation was found in the duration of the acquired sensitivity.

The first positive reaction in the skin was obtained as early as 1¼ hours, and the last positive reaction, as late as twelve weeks after transfusion. The maximum reactions occurred within one to five days and remained at this level as long as twenty-four days. Conjunctival and nasal responses came on as early as 2¾ and twenty-four hours, respectively, after transfusion. In the case of the eye, the last positive reaction occurred at the end of ten days, while the most prolonged nasal sensitivity lasted eleven days. After transfusion two of the patients developed constitutional symptoms (pruritus, asthmatic breathing) suggestive of responses seen in hypersensitive persons who have received an overdosage of antigen.

Chronic Clinical Allergy. Calcium and Potassium Therapy. Sullivan, C. J.: J. Missouri M. A. **38**: 125, 1941.

Thirteen patients of a group of twenty-nine allergic subjects suffering from asthma, allergic rhinitis, or urticaria were placed on potassium therapy consisting of potassium chloride in the form of a 25 per cent aqueous solution taken well diluted with water. Four of these thirteen were also on a high-potassium, low-sodium, and low-calcium diet. Of this group of thirteen, only one showed improvement. The remaining sixteen were given calcium gluconate in the form of powder or tablets, and nine of these were also placed upon a high-calcium, low-sodium, and low-potassium diet. Three of these sixteen patients reported symptomatic relief following calcium therapy. The results discourage the use of potassium and calcium salts in chronic clinical allergy.

Histamine Tolerance and Anaphylactic Death in Sensitized Guinea Pigs. Karady, E. S.: J. Immunol. **41**: 1, 1941.

The minimum lethal dose of histamine (that dose which will kill 50 per cent of all animals injected) for guinea pigs was found to be 0.02 to 0.025 mg. per 100 Gm. by intracardiac injection, and 0.18 to 0.2 mg. per 100 Gm. by subcutaneous injection. Guinea pigs injected subcutaneously every other day for four to eight weeks, with slowly increasing doses of histamine (0.005 mg. per 100 Gm. to a maintenance dose of 0.1 mg. per 100 Gm.) were found to be more tolerant to histamine than before pretreatment. This increased tolerance to intracardiac injection amounted to about 100 per cent; subcutaneous injection increased the tolerance by

only about 50 per cent. Attempts to increase tolerance by the use of relatively large doses of histamine given for three to four weeks caused exhaustion of the animals and death in about 60 per cent of them.

Anaphylactic death in guinea pigs sensitized to horse serum was prevented in 75 to 90 per cent of the cases by pretreatment with histamine. The most effective results were obtained when pretreatment was started immediately before the sensitizing injection of horse serum and continued for four to eight weeks afterward. The question is raised as to whether or not the administration of histamine to the animals before sensitization merely prevents them from becoming sensitized. The author was unable to protect guinea pigs sensitized to egg white against anaphylactic death by pre- and posttreatment with histamine.

Immunology

UNDER THE DIRECTION OF MATTHEW WALZER, M.D., AND
MAX HARTEN, M.D., BROOKLYN

Antigenic Properties of Substituted Serum-Globulin. Haurowitz, F., Sarafian, K., and Schwerin, P.: J. Immunol. **40**: 391, 1941.

The authors report experiments performed in order to determine whether the species specificity of natural proteins may be attributed to definite chemical ''determinant'' groups.

An increasing number of chemical substituents, such as acid and basic azo groups, iodine, and acetyl groups, were introduced into the molecule of native serum pseudoglobulin, and their influence on species specificity was investigated.

The introduction of 100 or more azo groups per molecule of globulin, as well as the introduction of the same number of iodine atoms or N-acetyl groups, inhibited the specific precipitation of serum pseudoglobulin by its homologous antiserum, thus indicating the destruction of species specificity by these procedures. Azoglobulin, iodoglobulin, and acetylglobulin containing fewer than fifty substituting groups per molecule could be precipitated as well as native pseudoglobulin.

Further experiments indicated that the specificity of native pseudoglobulin is destroyed by both substitution of all tyrosine groups and acetylation of the free amino groups.

The authors conclude that the species specificity of pseudoglobulin and probably other natural proteins cannot be attributed to a single ''determinant chemical group'' but rather to a definite structural arrangement of tyrosine groups, free amino groups, and perhaps other groups on the surface of the protein molecule.

Attempts to Sensitize Monkeys to Scarlet Fever Antitoxin. Toomey, J. A., Garver, W. P., and Takacs, W. S.: Am. J. Dis. Child. **61**: 518, 1941.

Ten Macac mulatta monkeys, given large amounts of highly purified and refined scarlet fever antitoxin intravenously over long periods of time and at varying intervals, failed to become sensitized to horse serum. This was indicated by the absence of positive cutaneous reactions to horse serum, by the absence of precipitin formation, and by the absence of any systemic reactions following the administration of a shocking dose of horse serum.

Experiments to determine whether or not animals sensitized to horse serum could be shocked by the subsequent administration of refined scarlet fever antitoxin were inconclusive. Five animals, given injections of normal horse serum in amounts equivalent to 1 per cent of their body weight, all developed precipitin titers from

1:100 to 1:10,000. No correlation between the presence or absence of a positive cutaneous reaction to horse serum and the precipitin titer could be made. In most instances, the precipitin titer was not elevated by subsequent injections. In some animals, precipitins appeared in the blood stream after the fourth or fifth injection of horse serum, only to disappear after subsequent injections. Two animals, one with a precipitin titer of 1:100 and the other with a titer of 1:1,000, died with shock symptoms as a result of sensitization to horse serum following an injection of the specific antigen.

Blood Platelets in Anaphylaxis. Kopeloff, N., and Kopeloff, L. M.: J. Immunol. **40**: 4, 1941.

A striking reduction in the number of blood platelets was noted in sensitized Rhesus monkeys following the administration of shocking doses of specific antigen to these animals. In general, the diminution in blood platelets paralleled the degree of shock symptoms. No significant change in the blood platelet count was observed in monkeys injected intravenously with large amounts of Witte peptone or histamine phosphate. A marked decrease in blood platelets also occurred in actively sensitized guinea pigs shocked specifically with the related antigen. Histamine shock, however, had no effect on the blood platelets of these animals.

Rabbits, repeatedly injected with crystalline ovalbumin and shocked one week later, showed a moderate decrease in blood platelets although clinical evidences of shock were not apparent. Nonsensitized rabbits showed no decrease in blood platelets following treatment with a shocking dose of antigen.

The Effect of Histaminase on the Gastric Secretory Response to Histamine. Atkinson, A. J., Ivy, A. C., and Bass, V.: Am. J. Physiol. **132**: 1, 1941.

The intramuscular injection of 8 to 15 units of histaminase in dogs with a Heidenhain pouch or with a pouch of the entire stomach failed to decrease the gastric secretory response of these animals to 0.5 mg. of histamine dihydrochloride. Previous studies had demonstrated that the continuous intravenous administration of histaminase during the absorption of histamine from subcutaneous tissue of dogs was also ineffective in decreasing the gastric secretory response. In sixteen human subjects the introduction of 100 units of histaminase into the duodenum, twenty minutes prior to the subcutaneous injection of 0.5 mg. of histaminase, did not decrease the gastric response. The ingestion of 50 units of histaminase, in the form of enteric coated tablets, for three days by eight human subjects also had no effect on their gastric secretory response.

The authors have been unable to demonstrate in this and in preceding studies that histaminase given by any route has an inhibitory effect on the gastric secretory response to 0.5 mg. of histamine.

Technic for Demonstrating Antibodies Against Tuberculin in Experimental Animals With Sensitized Collodion Pellets. Weir, J. M.: Proc. Soc. Exper. Biol. & Med. **48**: 47, 1941.

A method is described for utilizing suspensions of sensitized collodion pellets, following the technique described by Cannon and Marshall, for the study of antibody formation against tuberculin in animals immunized with killed tubercle bacilli.

Suspensions of collodion pellets sensitized with old tuberculin uniformly demonstrated the presence of antibodies against tuberculin in the blood serum of rabbits which had received injections of killed tubercle bacilli. A rise in

antibody production with increasing intervals of time after inoculation was noted. All of the preinoculation sera failed to cause agglutination of the sensitized pellets, as did control tests with saline and unsensitized pellets. Control tests on ninety sera from other laboratory animals confirmed the observation that this technique differentiates the sera of normal animals from those obtained from animals infected with tubercle bacilli. This method demonstrated the presence of antibodies against tuberculin in many of the animals at a time when complement-fixing antibodies were not demonstrable and at a time when many of the animals failed to exhibit cutaneous sensitivity to tuberculin. There was no demonstrable difference in the degree of antibody protection developed by animals injected intracutaneously with suspensions in saline and those injected subcutaneously with suspensions in paraffin oil.

The results of studies on human sera with this technique are now being analyzed.

Failure of Histaminase to Prevent Anaphylactic or Histamine Shock in Guinea Pigs. Youngner, J. S., Freedman, S. I., and Nungester, W. J.: Proc. Soc. Exper. Biol. & Med. **46**: 187, 1941.

Histaminase injected intracardially fifteen minutes or twenty-four hours prior to the administration of the intravenous shocking dose of the specific antigen did not inhibit or alter the anaphylactic response of sensitized guinea pigs. The production of histamine shock in normal guinea pigs was not altered by pretreatment with varying amounts of histaminase at different intervals before the intracardial injection of the histamine. Although no evidence was obtained of the in vivo activity of histaminase in the above experiments, the authors believe that there is still a possibility that histaminase might act in vivo if histamine were released slowly and in small amounts, thus permitting more time for the enzyme to act.

Studies With Colored Antigens. I. The Rate of Disappearance of Dye-Albumin From the Blood. Pratt, H. N., and Gregersen, M. I.: J. Immunol. **40**: 163, 1941.

The conjugation of a dye, R-salt, with recrystallized egg albumen altered the specificity of the original albumin. This conjugated dye protein (R-salt-azo-benzidine-azo-egg albumen) was injected intravenously into rabbits and a dog, and the rate of its disappearance from the blood plasma was determined by spectrophotometric determinations. The dye protein disappeared rapidly from the plasma, 30 to 80 per cent escaping during the first hour. The dye protein was completely eliminated from the plasma in twelve to twenty-four hours, depending upon the amount injected.

Only five per cent or less of the dye protein was withdrawn from the plasma by the blood cells. None could be detected in the urine by colorimetric or immunologic methods. The rapid removal of this protein from the plasma cannot be explained either by a reaction between it and the cellular elements of the blood or by excretion through the kidney.

The in Vitro Action of Trichina Larvae in Immune Serum—a New Precipitin Test in Trichinosis. Roth, H.: Acta path. et microbiol. Scandinav. **18**: 160, 1941.

A new precipitin test in trichinosis is described. About 100 living trichina larvae, obtained from muscles of rabbits or guinea pigs at least eight weeks after infestation, are placed in the depression of a sterile hollow-ground slide and covered with 0.5 ml. of sterile, unheated serum. The cover-slip is rimmed with vaseline. Such preparations are incubated and examined microscopically at intervals.

Twelve sera of heavily infected guinea pigs, seven of infected rabbits, and twenty-six human sera from patients suffering from clinical trichinosis were tested. After from three to six hours, distinct bubble-like precipitates could be seen microscopically at the anterior ends of many of the larvae incubated in the immune serum. These precipitates increased in size during the following hours. After from sixteen to twenty-four hours the serum contained quantities of free, cast-off precipitates. No specific deleterious effect of the immune sera or of the microscopic precipitates on the larvae was observed.

A comparison of the usual Bachman precipitin test with this microscopic test employing living trichina showed a definite parallelism in results. It is suggested that the microscopic precipitin test may be of greater aid in early cases or in those cases in which only a very slight antibody production has taken place.

Pediatrics

UNDER THE DIRECTION OF WILLIAM C. DEAMER, M.D., SAN FRANCISCO

Allergic Eczema of Infancy and Childhood: Application of Skin Testing. Miller, H., Piness, G., and Small, W.: California & West. Med. **54**: 267, 1941.

Skin testing using the scratch or puncture method first, then the intradermal method if necessary, is looked upon as a highly efficient method of diagnosis and control of allergic eczema. The statement is true only if the diagnosis is correct, the tests properly done, their limitations appreciated, and the results of tests intelligently applied.

Extensive lichenification, fungus infection, pyoderma, contact dermatitis, and dermatitis *medicamentosa* complicate treatment and retard healing. Forty per cent of the cases in which the etiology was known were caused by foods alone; 20 per cent, by pollen alone; 11 per cent, by epidermals alone; and the remaining 29 per cent, by foods, pollens, and epidermals in combination. Five per cent of the cases of allergic eczema gave completely negative skin tests even on repeated testing.

Immediate Allergic Reactions to the Schick Test. Beck, J. R.: Delaware State M. J. **13**: 26, 1941.

Three cases of immediate allergic reaction to the Schick test are reported.

All reactions occurred in older children who had previously been Schick tested one or more times without any unusual reaction. All had received alum precipitated toxoid, and two had also received plain toxoid years before. The reactions all occurred within twenty minutes and involved a generalized erythema and some urticaria and swelling of the face; two of the children were nauseated. There was no immediate family history of allergy in any of the cases. Two patients had received their alum toxoid at the same time and presumably from the same source.

Through an oversight the Schick test was repeated in one instance and an intensified general reaction of the same type followed. Adrenalin was used with prompt benefit in all three children.

The Management of Eczema of Infants and Small Children in the Hands of the Practical Doctor. Lauenstein, H.: Deutsche med. Wchnschr. **67**: 353, 1941.

The first rule is to take cow's milk or goat's milk out of the diet. Buttermilk is used instead, and in addition, bouillon, vegetables, banana purée, and scraped

apple. As medication three drops of Fowler's solution three times a day are used; this treatment undoubtedly softens the skin. More important, however, is the external care of the skin. Bandages and masks are indispensable. The skin is kept covered by them for at least eight weeks, even though improvement may be apparent sooner than this. Coal tar is not used; boric ointment has largely replaced it and zinc preparations. Thus, the skin is protected from external irritants such as cold, water, air, and above all sunshine. Although the parents fear the sudden death of eczema patients and the occurrence of asthma, the author has not had the latter complication when this treatment has been given. A recent valuable additional help would seem to be chemotherapy with the sulfonamides. Eubasin was found to be much superior to prontosil and was given by mouth in small daily doses for five days, followed by two days of rest.

Investigation of the Skin Reaction to Bee Venom in Allergic Diseases. Traub, E.: Acta paediat. 27: 177, 1939.

On theoretic grounds bee venom seemed to be a suitable material for determining a nonspecific parallergic effect on skin responsiveness in certain diseases. It was thought that the study of the skin reaction to the administration of bee venom intracutaneously would throw some light on the changing allergic status of the patient, quite apart from specific antigen-antibody reactions. The test consisted in injecting 0.06 c.c. of 1.5 per cent solution of bee venom.

In bronchial asthma, large-sized reactions were observed. In tuberculosis the reaction roughly paralleled that to tuberculin. In catarrhal jaundice reactions of increased size were obtained.

Small reactions occurred in croupous pneumonia and early in scarlet fever. In the latter disease, a blanching effect similar to the Schultz-Charlton reaction was observed. In rheumatic fever a decrease in the size of the reaction occurred at the time of rheumatic rashes such as erythema *annulare* and urticaria *rheumatica*.

Rhinology and Ophthalmology

UNDER THE DIRECTION OF FRENCH K. HANSEL, M.D., ST. LOUIS

Sodium Morrhuate Therapy for Chronic Simple, Vasomotor and Hypertrophic Forms of Rhinitis. Thacker, E. A., and Hauser, G. H.: Arch. Otolaryng. 33: 600, 1941.

Thacker and Hauser report their observations on the use of sodium morrhuate for chronic simple, vasomotor and hypertrophic forms of rhinitis. This form of therapy is recommended in those cases of chronic nasal obstruction with engorged turbinates which have failed to respond to the usual types of therapy after two months' trial. This type of treatment is not advised for cases of definite allergy with nasal manifestations which can be controlled adequately by desensitization or removal of the allergens. Nasal obstruction and associated symptoms were relieved completely in 66 per cent of the patients. The remaining 34 per cent were entirely or considerably relieved of nasal obstruction, and their postnasal drip and headaches were definitely diminished. The number of injections of a 5 per cent solution of sodium morrhuate solution to produce the desired effect varied. Seventy-two and four-tenths per cent of the patients required only one or two injections in one or both inferior turbinates. The length of time this treatment will relieve the symptoms has not been ascertained. Eight months have elapsed since the first patients of the series received their last injection. So far, none of the patients has had

a recurrence of nasal obstruction, and there has been no clinical evidence of any deleterious effect of the drug on the nasal mucous membrane. These investigations are to be continued for another year.

The Use of Histamine in Ménière's Disease. Horton, B. T.: Surg., Gynec., & Obst. **72**: 417, 1941.

Forty-nine cases of Ménière's syndrome were treated by the intravenous injection of histamine and obtained prompt relief. The dosage employed was 1 mg. of histamine base (2.75 mg. of histamine diphosphate) in 250 c.c. of physiologic saline solution. This was administered intravenously by the gravity method at the rate of fifty or sixty drops per minute. One and one-half hours were usually required for the treatment. Injections may be repeated on successive days. To prevent future attacks, a maintenance dose of from 0.1 to 0.2 mg. of histamine was given subcutaneously from two to four times a week.

Roentgen Ray Therapy for Patients With Chronic Sinusitis. Youngs, N. A.: Arch. Otolaryng. **33**: 550, 1941.

In a series of seventy-five cases Youngs evaluates the efficacy of roentgen ray therapy for chronic sinusitis. Many of those treated, he states, were definitely known to be allergic, although no extensive studies of allergy were made. The effectiveness of the therapy normally could not be evaluated until a lapse of at least two to three months. No harmful effects were noted in any of the patients.

The best results were obtained in children, or patients under 17 years of age. Marked beneficial results were obtained in mildly allergic persons, but in the severely allergic no benefits were obtained.

An Evaluation of the Ménière Syndrome. Grove, W. E.: Ann. Otol., Rhin. & Laryng. **50**: 55, 1941.

In an evaluation of the Ménière syndrome presented by Grove, it is pointed out that, according to the reports of various observers, allergy plays some part in the etiology of this disease. It frequently appears in patients with other manifestations of allergy; the withdrawal of the offending allergens will cause a cessation of the attacks and the reintroduction of the allergen will again produce an attack. Vogel noted that among eighty-three patients with the Ménière syndrome, five had hay fever, nine had urticaria, and fifteen had nasal allergy and five asthma, an incidence of allergy of about 40 per cent. It was further stated that the localization of the trouble is not in the nerve alone because no pathology has ever been demonstrated there microscopically.

Headache was found by Dederding to be the most frequent extra-aural symptom, being present in 91 per cent of a series of 228 cases. It has been pointed out by a number of observers that water retention is associated with the attacks. This is manifested consequently by an increase in weight and thereby suggests the presence of angioneurotic edema.

Symptoms of Gastrointestinal Origin in the Ear, Nose and Throat. Gatewood, W. L.: Arch. Otolaryng. **33**: 592, 1941.

In discussing the symptoms of gastrointestinal origin in the ear, nose, and throat, Gatewood calls attention to the possible association of gastrointestinal allergy and nasal allergy. Among the symptoms in the ear, nose, and throat the following were enumerated: postnasal drip, vertigo, fullness in the ears, rhinorrhea, recurring

colds, cough, dryness and stuffiness of the nose, and headache. Five case reports were presented to illustrate the main points in the presentation.

The otolaryngologist not infrequently observes patients with nasal allergy who also have definite gastrointestinal symptoms from food. Sometimes the internist overlooks the allergic nature of the gastrointestinal symptoms in these cases.

Nasal Obstruction: Is It Allergic? Oppegaard, C. L.: Minnesota Med. **23**: 112, 1940.

Oppegaard believes that every case of nasal obstruction, either acute or chronic, should be considered as of possible allergic origin until proved otherwise. It is pointed out that often the patient who has a marked deflection of the septum may not complain of nasal obstruction. Oppegaard states that in his experience, 30 per cent of the cases with nasal pathology present local findings attributable to allergy or are associated with other manifestations of it. Emphasis is also placed upon the importance of recognizing allergy as the cause of nasal obstruction in children. Allergy, he states, explains many poor results obtained from removal of the tonsils and adenoids in children.

Correspondence

The Editor,

THE JOURNAL OF ALLERGY.

On Oct. 1, 1941, there will be a vacancy at the New York Post-Graduate Hospital in the position of a full-time research fellow in allergy, available to graduates of accepted medical schools, who have completed a two-year internship in an approved hospital. An honorarium is provided.

For further particulars, communicate with W. C. Spain, M.D., 116 East 53rd Street, New York, N. Y.

W. C. SPAIN

June 5, 1941

Erratum

In the May issue of the JOURNAL in the article "Studies on Blood Histamine in Cases of Allergy" by Bram Rose, in the last paragraph on page 328 the phrase "with variations of from 2.0 to 75.0 y/100 c.c. (0.02 to 0.75 y/c.c.)" should read: "with variations of from 2.0 to 7.5 y/100 c.c. (0.02 to 0.075 y/c.c.)."